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*Blood Pressure, Report of Joint Committee on Mortality of The Association of Life Insurance Medical Directors and The Actuarial Society of America. New York, 1925. pp. 10, 52.

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THE AMERICAN JOURNAL
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ORIGINAL ARTICLES

SPECIAL METHODS FOR THE DIAGNOSIS OF TUMORS IN THE LABORATORY

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It should be made quite clear at the outset of this paper that there is no royal road to the diagnosis of tumors in the laboratory; whatever methods be used to differentiate oneneoplasm from another the diagnosis still depends upon the training and experience of the observer. Tumors have definite physiognomies and you all became more or less familiar with these in the 2nd year classroom, when you studied carefully selected sections or specimens. Most of you have, I fear, stored this somewhat painfully acquired knowledge on the top shelf and, should you take it down and look it over, you would find that it had accumulated a lot of cobwebs.

The diagnosis of tumors is largely a matter of seeing many examples of them and, just as one becomes acquainted with people, getting to know them without having to turn on the high-powered lens and search for details. There is no stain that will automatically issue a diagnosis and there are few rules in the recognition of tumors that do not have their exceptions. A few weeks of studying sections in a laboratory of surgical pathology is worth many months of reading in textbooks. Some tumors have many variants or differing types of growth, no one type may be considered to be the inflexible prototype of that particular neoplasm.

Granted that the pathologist has the necessary background, however, there are many methods of going about the securing of a diagnosis, some of them partial or indicative and useful for screening out obvious cases, others ancillary to the routine, but in the end the paraffin section, properly stained, is the last court of appeals.

In presenting this subject it would be well to begin with the simpler methods and progress to the more complicated procedures. Simple methods are calculated to cause the least disturbance to the patient, physician, surgeon and pathologist. Those which are more complicated often produce discomfort for the patient and make demands upon the skill and experience of the physicians involved.

Smears. The recent work of Papanicolaou has given great impetus to the diagnosis of malignant growths in smears. He began by studying the cytologic changes in the vaginal epithelium in vari-

* Delivered at the University of Buffalo, School of Medicine, Centennial Symposium on Cancer, October 1, 1946.

ous phases of the menstrual cycle and under varying conditions of hormonal stimulation. While carrying out his investigations he noted that cells of malignant tumors were encountered in some of the smears. As these were readily recognized he accordingly expanded his technique to facilitate this form of diagnosis. At first a side line in his investigation, this very important feature has become a main line and he has devised methods for examining not only vaginal smears, but also material obtained directly by intrauterine aspiration. Thus he was enabled to narrow down his sources of pathologic cells and to determine definitely that a given tumor was intrauterine and not located in the cervix uteri.

It was not long before he branched out and began examining smears of other fluids and secretions. It was found that the urine from a bladder which was the site of carcinoma might contain carcinomatous elements and that these could be concentrated by centrifugation and thus readily identified. A noteworthy example of the practical value of this method was the discovery of cells from a carcinoma in the urine of a bladder which, on cystoscopic examination, showed no carcinoma. Investigation revealed a fairly large diverticulum and, at operation, a carcinoma was found tucked away in the fundus of this, safely hidden from the visual field of the cystoscope.

It was evident from the success of these 2 lines of experimentation that the method might be extended to an examination of aspirated fluids from other hollow viscera such as the stomach and the pulmonary tree. Accordingly these were investigated and it soon became evident that the method was of distinct value in connection with the examination of smears of gastric contents and of sputum or bronchial washings, although diagnosis was found to be more difficult in this case than in that of the genitourinary system.

The success of the method depends upon the fact that carcinomas tend to break down superficially and to cast off cells into the fluid that bathes them. If the malignant tumor be intramural and does not desquamate into the cavity of the organ, the method will, of course, not succeed. This would be true of leiomyoma of the wall of a viscus, as well as of leiomyosarcoma—unless that tumor had eroded its way through the lining mucosa.

There is nothing miraculous about the technique; it depends simply upon 2 factors: carcinomatous cells must be present and the pathologist who searches for them must have had sufficient experience to be able to recognize them when he sees them. The procedure, then, should not be entrusted to anyone who is unfamiliar with cancerous elements, but should be carried out by an experienced pathologist. The qualified pathologist is usually well acquainted with the appearance of carcinomatous or sarcomatous cells; they are usually of abnormal size, irregular outline and poor histologic differentiation; they are generally very variable in diameter; they present nuclei that are hyperchromatic and stain intensely, often possessing very large and prominent nucleoli. They may show normal, or very abnormal mitotic figures. Thus it becomes easy for an experienced observer to recognize them and report their presence in a given smear.

The method has few drawbacks. A carcinoma of any size will usually cast off cells or clumps of cells which may be found after a few minutes of searching. If the pathologist is very much busied with routine, it is not often feasible for him to spend $\frac{1}{2}$ hour or so in searching through a smear poor in cellular elements. A few such smears would occupy an 8 hour day! It is found, however, that if carcinoma is present, its cells are usually so striking in their appearance that they are readily found after a few minutes.

One needs very little equipment for carrying out such examinations. The fluid is either smeared-out as it comes to the laboratory, without concentration, or if it be very dilute (like urine or bronchial

washings) it may be necessary to concentrate it by sedimentation. The smears are allowed to dry on microscopic slides and are then fixed in equal amounts of ether and alcohol. They are then stained by some good method like the trichrome stain of Masson. The cellular details as revealed by this are excellent, and one gains very beautiful pictures of ciliated cells from normal bronchial mucosa, the cilia bright red and the cytoplasm grayish green. The carcinomatous elements stand out by their lack of differentiation and their rather drab color reaction. When urine is to be examined, Papanicolaou and Marshall have found it advisable to mix about 40 cc. of this in a test-tube with 10 to 20 cc. of 95% alcohol. This is then centrifugated and the sediment smeared onto glass slides and fixed in alcohol and ether. It should not be forgotten that prostatic fluid is excellently adapted to examination in smears, it may be expressed by massage, smeared out and fixed in alcohol and ether, and then stained and examined for cancerous cells.

Smears from the alimentary tract are not as yet very satisfactory on account of the distortion of cells by digestion and of the presence of many foreign, though normal cells, from ingested meat and the like. For the present, direct biopsies appear to be preferable in this case.

Aspiration Biopsies. These fall into 2 categories: the material aspirated in the form of serous effusions from the pleura, pericardium or peritoneum, or the cavities of cysts or joints; and, secondly, solid matter aspirated through a needle or cannula attached to a syringe.

Serous Fluids. The examination of serous fluids for cells of malignant tumors is facilitated by allowing them to sediment by gravity in the refrigerator overnight and then centrifugating such sediments mechanically, after discarding the supernatant fluid and replacing it with a fixative such as formalin, Zenker's or another such mixture. The button is then carefully removed from the bottom of the tube and embedded in paraffin, in which it is sectioned exactly like a piece of tissue. Such sediments may be stained as one may desire. It will be found that the pathologist must acquire a certain degree of familiarity with those cells normally present in transudates and exudates in order to recognize the abnormal elements. The chief difficulty is encountered in connection with desquamated mesothelial elements from the pleura, pericardium, or peritoneum; these may often take on very substantial proportions and mislead the pathologist. In hepatic cirrhosis there is usually a massive proliferation and desquamation of peritoneal cells, but these are remarkably uniform in size, shape and appearance, while the reverse is true in the case of neoplastic elements, which vary conspicuously in these respects. The presence of multinuclear cells and of mitotic figures in mesothelial cells is often confusing to the beginner, but as the effusion constitutes an excellent culture medium and the cells multiply and live in this much as would bacteria in broth, it is not surprising that they should take on a quasi-neoplastic appearance. When cancer is present one may find masses of neoplastic cells, sometimes attached to stromal elements, in the sediment. These groups may often be so characteristic in their appearance and arrangement that one may make a fairly accurate surmise as to the type of tumor and its primary site. The method is decried in some quarters, but this is unjust, as it has a very distinct value which should not be underestimated; in our laboratory the positive reports are better than 85% accurate, which speaks for itself.

Aspiration of Solid Neoplastic Tissue. The "needle biopsy" has enjoyed considerable vogue for a long time and consists in introducing a coarse aspirating needle, or cannula, into the depths of a suspected neoplasm and attempting to aspirate some of this into the attached syringe by exerting strong suction while, at the same time, oscillating the tip of the needle to dis-

lodge bits of tissue and facilitate their withdrawal. Oftentimes valuable fragments are thus detached and can be smeared out onto a glass slide, or even embedded in paraffin and sectioned. The method is frequently employed in our hospital for exploring the liver, even in cases where there is no suspicion of the presence of carcinoma and all that is wanted is a diagnosis of the hepatic parenchyma. Aspirated material should be fixed in an appropriate medium before it is stained. We used to emply heat, simply smearing aspirated tissue onto a slide and passing the latter through a Bunsen flame. This does not give satisfactory results, as the procedure is too drastic and the cellular details are obscured by violent coagulation. Fixation of the smear, after it is dry, in equal parts of alcohol and ether, is now our method of choice.

The aspiration biopsy has definite disadvantages: it does not give one enough tissue to afford any concrete idea of the topography of a given tumor, it merely produces its cells; it often fails entirely in the case of dense fibrous growths, where the cells refuse to become dissociated from their fellows and one obtains merely an excellent smear of blood. It has always seemed, after long experience, that it is far better to excise a bit of tissue for a biopsy and thus disturb a potentially malignant tumor as little as possible. Agitating a needle in its depths may detach cells for diagnosis, but it might also set them free into the circulation and favor metastasis. On the other hand, a piece of tumor that is cleanly excised with a sharp scalpel would seem to suffer far less cellular dissociation and thus tend to present less likelihood of metastasis. This is, of course, theoretical but I believe that it is a valid point.

Tissue Biopsy. There is little to be said here about the biopsy of tissue for diagnosis, as its excision is a self-explanatory matter and the treatment of excised tissue differs not a whit from that of a tumor which has been excised *in toto*. It is, however, advisable to stress the necessity for the exercise on the part of the surgeon of a reasonable amount of discernment in the choice of the tissue he will excise for examination. The pathologist can diagnose only that which is in his specimen; if the surgeon cut out a piece of tissue near a tumor, rather than in it, he may miss the tumor entirely and the pathologist, failing to find any neoplastic tissue will, of necessity, report its absence. If the surgeon exposes a mammary tumor, for example, and then cuts out a piece of tissue that is hard and brawny he may get the important part of the growth; if he selects a piece that is stony-hard and which, when incised with the knife, cuts like balsa wood and leaves the edges of the incision sharply angular and not rounding-off, the matter will be simpler. If, on making the section, he notes yellowish streaks on the sectioned surface, or the yellowish "worm casts" of a comedocarcinoma, he will be even more surely producing "pay dirt" for the pathologist. A biopsy should always be as generous as is feasible, tiny bits of tissue are hard to handle in the laboratory, they are difficult to keep track of in the solutions used and they are little better than aspiration biopsies in respect to the fields of microscopic observation that they afford.

We should consider frozen sections. Surgeons should see a few of these made, if they have not already done so. They will then realize the futility of expecting a pathologist to make such sections on biopsies measurable in millimeters only, or consisting of feathery, papillary growths that fly apart after they have been frozen and cut and floated on water. All that holds a frozen section together is the tissue itself, once the ice that serves as the impregnating medium, has melted. Ice is not at all similar to paraffin or celloidin! Uterine curettings, unless very bulky and fleshy, are extremely difficult to manage on the freezing microtome; it is much better to allow the pathologist time to

run such material through in paraffin by one of the more rapid methods. It has at least once happened that I have been requested to make a frozen section of a bony tumor! A little thought should convince the operator that bone requires decalcification before it can be cut and that decalcification is a matter of days.

Special Stains in Diagnosis. The average American pathologist is a man who is used to thinking in terms of red and blue; the nuclei of the cells in a section stained by the usual hematoxylin-eosin, or eosin-methylene blue methods are blue, everything else is red. This is all very well for the diagnosis of simple, "run of the mine" sections from familiar tumors; but when a growth is complicated, this simple and overworked method falls down lamentably. It is strange that so many pathologists depend upon it exclusively and, when they send sections of tumors for consultative diagnosis (tumors that are thus admittedly tricky), send only those stained by this conventional technique. The advantage of a colored map of, say, the State of New York, with the counties in contrasting colors, is the fact that it is much more rapidly understood than one in black-and-white; this is no less true of microscopic sections in which various tissues assume various colors in the slightly more complicated stains, but only 2 in hematoxylin and eosin.

Thus a good polychrome stain is vastly superior because it affords immediate orientation as to the various components of the tissue to be examined and time is not lost in trying to identify these elements by morphology alone. As connective tissue fibers are stained green or blue in Masson sections, while muscle is red and neural sheaths golden yellow, one wastes no time in identifying them. Thus a fibroma would be mainly green and a muscular tumor mainly red, erythrocytes would be orange, fibrin vermillion, keratin orange and mucinous tissue green.

There is no facile formula for the diagnosis of a malignant as opposed to a non-malignant tumor insofar as specific stains are concerned; it is purely a matter of morphology and familiarity with the criteria already adumbrated elsewhere in this paper. When it comes to identifying the source of a tumor, however, the matter is different. We have at our disposal an enormous number of stains for the demonstration of specific chemical substances and histologic elements which enable a reasonably versatile pathologist to come very close to an accurate determination of the tissue-of-origin of a given growth and this may often be of great importance as it may indicate wide differences in the prognosis.

Let us suppose that a tumor is full of granules of a brown pigment: is this melanin, or is it not? If it be melanin, the prognosis may be very bad. Recourse to Perl's stain, which is merely the old Prussian blue reaction for free iron, will at once determine whether the pigment is hemosiderin. This is a common by-product of degenerated hemorrhage and means little in the prognosis. If the granules turn greenish blue the pigment is hemosiderin, if they remain brown it is something else. Impregnation with appropriate solutions of silver salts will turn melanin black, even in its colorless, or premelanic phase.

The prognosis of mammary carcinoma is said to be better if mucous deposits are found within the cells, or in the stroma. This substance is readily recognizable without special stains in the latter case, but when it is included in the cellular cytoplasm it escapes notice in ordinary sections. In those stained with Meyer's mucicarmine,* however, each droplet of mucus will come out bright carmine and one may readily determine the general prevalence of mucus in the section and render an accordingly more favorable prognosis. This has worked out well in our laboratory. Another carmine stain is used to demonstrate glycogen and there are several tumors which contain goodly amounts of this, notably the renal celled

* The Dresbach modification has been found to be excellent (Dresbach, M., Arch. Path., **44**, 391, 1947).

carcinoma of the kidney, formerly called "hypernephroma" and the rare chordoma, which develops from rests of the notochord. It is, however, seldom necessary to demonstrate glycogen in order to make a diagnosis of these neoplasms.

Rhabdomyosarcomas are fairly common and very often misdiagnosed. In establishing an iron-clad diagnosis it is desirable to demonstrate cross-striations in the cells of these growths; this is best accomplished by using Mallory's phosphotungstic acid hematoxylin, a stain that is far simpler than its rather formidable name. Cross-striations develop relatively late in the embryologic development of muscle and it is not always possible to demonstrate them in rhabdomyosarcomas; one should always endeavor to do this, however.

Leiomyosarcomas may be difficult to identify unless one employs impregnation of the reticulum with silver and demonstrates the parallel fibrils of sarcolemma that enmesh the cellular elements. These tumors sometimes resemble carcinomas, particularly when they arise in the muscle of the gastric cardia. A trichrome stain *á la Masson* will bring out the sarcoplasm of component cells as bright red, polar accumulations, thus differentiating them from epithelium or fibroblasts.

One of the knottiest problems in the diagnosis of tumors exists in connection with the group of lymphosarcomas and Hodgkin's granuloma. Here it is imperative to employ Zenker fixation and the Giemsa stain, which has proved its worth in connection with lymphoid tissue. The eosinophils which constitute one-third of the "Hodgkin's trilogy" are brought out in bold relief by this method, the lymphoid elements are more specifically stained and their basophilic qualities accentuated. Thus the latter can be differentiated from the paler reticulum cells.

Those of us who put store in separating the tumors of the reticuloendothelium (or "retothelium," as Roulet calls it) from the other elements of lymphoid tissue will find silver impregnation invaluable in clinching the diagnosis. The fibrils of reticulum penetrate among and around the cells of the tumor and thus differentiate it from a carcinoma, which it sometimes resembles. In carcinomas the reticulum forms a basal membrane about groups of epithelial cells and does not penetrate into their complexes at all conspicuously.

The tumors of the nervous system, whether central or peripheral, have only comparatively recently been placed upon a scientifically devised basis by means of recourse to silver impregnations of various sorts, depending principally upon the methods of Ramon y Cajal, who employed silver nitrate, and on those of Bielchowsky, who used a double salt of silver, the diammino hydroxide. A large number of methods were derived from these by del Rio Hortega, Bailey, Penfield, Globus and many other investigators. By using these procedures it is possible to split up the large groups of "gliomas" into a number of subsidiary types each of which carries a slightly different prognosis. The old diagnosis of "glioma" was unpredictable, the modern classification ranges from the non-malignant astrocytomas and oligodendrogliomas to the very malignant glioblastoma multiforme, which is unfortunately among the commonest of cerebral tumors. Once a pathologist has familiarized himself with these forms of neurogenous tumor, the invariable use of silver impregnations becomes relatively unnecessary; a good Masson trichome stain is usually sufficient to present evidence which may be accurately evaluated and diagnosed, although it is usually followed up by silver impregnations (which take more time to prepare) as a confirmatory measure.

Tissue Cultures. Stout and his collaborator Murray have recently shown the advantages of growing tumors in tissue cultures; this is a very specialized technique which is delicate and rather capricious until mastered. It began with Harrison's experiments with the explanted amphibian nerves and grew under the direction of Burrows, Carrel, Ebling and

others until it attained universal attention and was also extensively used in England and Germany. Stout and Murray have been explanting small pieces of human tumors into appropriate media and have studied the individual cells as they grew out from the explant. In this way they have been able to observe differences between fibroblasts and lemmoblasts and to determine that a neurofibroma is really a rather scirrhous form of neurilemmoma, most familiar to you as the so-called "acoustic neurinoma." Their experiments are not confined to neurogenous tumors, as they have subjected many types of growth to test in tissue cultures.

Akin to the tissue culture is the study of tumors explanted into the anterior chamber of the eyes of experimental animals. Greene has done very valuable work along this line. While the transplantation of tumors from one species to another usually fails if the explant is placed in subcutaneous tissue, muscle, or a body cavity, the anterior chamber of the eye appears to be incapable of producing antibodies that destroy such explants in these other situations. Thus the growth of a tumor may be studied through the slit-lamp microscope as it progresses in the transparent organ. It takes these explants a long time to become accustomed to their new habitat and to grow there, but once they begin to grow they make rapid progress. Both the tissue culture and the ocular culture are more suited to experimental observation than to diagnosis, but it is only by experiment that we get to know something about these tumors.

Laboratory Procedures in Diagnosis. As certain neoplasms cause changes in the biochemistry of the body, this fact has been employed as a diagnostic adjuvant in the case of such growths. Choriocarcinoma will increase the chorionic gonadotropin (formerly called "prolan") of the blood and urine, so that the patient reacts to its presence as would a pregnant woman to that of a placenta. Carcinoma of the prostate is accompanied by increase in the acid phosphatase content of the blood, particularly when there has been widespread metastasis. Bence-Jones proteins appear in the urine of patients with multiple myeloma of bone, as well as in the presence of some other tumors. Desoxycorticosterones are increased in the case of cortical adenomas of the suprarenal. All these phenomena, when proved, are merely indicative of the nature of a tumor; its final diagnosis will nevertheless still depend upon its appearance under the microscope.

For a while it was fashionable to employ certain fish to serve as indicators of increased gonadotropin in the urine. A little of this liquid from a pregnant woman, or a patient suffering from choriocarcinoma or malignant melanoma, when added to water in which these fish were kept would cause them to react characteristically. The male of the German minnow known as the "Elritzer" would take on gorgeous colors along his belly, the female bitterling would protrude her ovipositor, the female African frog would immediately lay eggs if gonadotropic material was present. Naturally the Aschheim-Zondek test is also applicable in such cases and has outlived the other methods in usefulness. But these tests are merely confirmation of the fact that there is something present in the patient which is producing gonadotropic hormones and it will not diagnose the tumor. While melanomas will bring about a positive reaction in tests for gonadotropin, it has been found that neurogenous sarcomas will also do this. There has, as yet, been no real need for the hospitals to dispense with their histopathologists, or for the latter to discard their microscopes[8].

At one time the Gordon test for Hodgkin's disease gave some promise of diagnosing that condition without resorting to biopsies of tissue, beyond removing small fragments for injection into rabbits. In the presence of Hodgkin's disease the

rabbits would develop characteristic paralysis. Later experimentation proved that this was attributable to the presence of eosinophils in the tissue and that any tissue richly infiltrated with them would provoke a positive reaction; thus the test was found to be non-specific and has been largely given up.

The functional action of tumors has been mentioned already; islet tumors of the pancreas will provoke hyperinsulinism, medullary adenoma of the suprarenal will bring about hypertension, cortical adenomas of the same gland will produce precocious sexual development and growth in children, masculinization in the female and less marked sexual changes in the male. Tumors of the parathyroid occasion changes in the calcium metabolism of the body and in the histology of the bony skeleton. All these can be more or less accurately checked up by assays or by clinical tests, but the actual diagnosis still rests upon the outcome of histopathologic observation.

Summary. This paper, when summed up, will merely indicate that there are no "special tests" or methods for establishing the diagnosis of malignant tumors in the sense of infallible and simple procedures that are comparable with clinical pathologic assays, serologic complement fixations and the like. We are still largely dependent upon the gross and microscopic examination of the tumors, or samples of them, under the microscope. One may speak of special methods, then, only insofar as they are specialized procedures in "tissue pathology."

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FOLIC ACID THERAPY

RESULTS OF A CLINICAL STUDY*

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IT is the purpose of this paper to report the results of the administration of folic acid in a variety of hemopoietic disorders. Included in this study are cases of: Addisonian pernicious anemia, tropical sprue, macrocytic anemia of unexplained origin, anemia of renal insufficiency, leukemia, idiopathic leukopenia, irradiation leukopenia associated with malignancy and leukopenic states of Addison's disease, Felty's syndrome and thiouracil intoxication. Thirty-four cases in all have been studied.

Not since the introduction of liver treatment for pernicious anemia by Minot and Murphy in 1926[16] following the experimental work of Whipple[23] has so much attention been focused upon potent antianemic substances. The clinical and experimental significance of folic acid, particularly, has contributed to our knowledge of pernicious anemia.

Folic acid, so named by Mitchell, Snell and Williams[17] because it may be extracted from spinach, is a recently recognized member of the vitamin B complex. As is so often the case, the credit for the discovery of this drug cannot be assigned to any one individual, but rather to many different groups of investigators who were working in highly diversified fields.

One of the first and most important contributions to the study of the effect of vitamin deficiency diseases upon hemopoiesis was that made by Wills, who in 1931[24] noted the striking effect of marmite, a yeast extract, on the blood picture of "tropical anemia" and "pernicious anemia of pregnancy." She related the hemogram of these conditions to Addisonian pernicious anemia and stated that these diseases differed only in symptomatology. She shrewdly observed that "tropical anemia" and "pernicious anemia of pregnancy" were deficiency diseases and were corrected by an unknown substance in marmite, probably not protein in nature. She stated that "Marmite is, however, known to be a rich source of the B vitamins." Later, in 1932 Wills and Bilimoria[25] carried out experiments in monkeys wherein it was shown that certain vitamin B substances could correct or cure the nutritional cytopenias of these animals. In 1935, Day, Langston and Shukers[5] were able to corroborate Wills' work on monkeys. The substance present in dried brewer's yeast which was instrumental in correcting the nutritional cytopenias of the monkey was designated vitamin M by Day, Langston and Darby[6] in 1938.

Likewise in 1938, Stokstad and Manning,[21] working on chicks fed all the known essentials, noted that they needed a substance present in yeast, middlings, alfalfa, or wheat bran for growth. In the next year, Hogan and Parrott[11,12] reported similar findings and added that chicks fed a diet lacking in this unknown factor also developed a "macrocytic hyperchromic anemia." This factor, present in liver, they called vitamin B_c.

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From the seemingly unrelated field of bacteriology came the next important advancement, for in 1940 Snell and Peterson[18] reported a substance, present in yeast and liver, which could be adsorbed on norite and eluted therefrom with ammoniacal alcohol. This substance was found to be necessary for the growth of *Lactobacillus casei* and *Streptococcus lactis*. It was called by them the "norite eluate factor."

In 1944 Hutchings, Stokstad, Bohonos and Slobodkin[13] reported further work on the "*L. casei* factor." This substance they related to vitamin M, vitamin B_c and the norite eluate factor.

In 1941 Mitchell, Snell and Williams[17] were able to secure a substance from spinach which was essential for the growth of *Strep. lactis* and was defined by them as a growth factor for this particular organism. It was called folic acid.

The last important contribution to our knowledge of folic acid (other than clinical contributions) was its synthesis in 1945 by Angier *et al.*[1] and finally the announcement of its chemical structure by this same group in 1946.[2] Table 1 is a brief summary of some of the literature dealing with folic acid.

was anemia. These patients were studied as follows: After proper baseline studies, including red blood cell count, hemoglobin, white blood cell count, reticulocyte count, examination of the peripheral blood, and hematocrit studies, the patient was placed on either oral or parenteral folic acid. Thereafter daily reticulocyte counts were done. White blood cell counts, hemoglobin and red blood cell counts were done twice weekly. Subsequent to the termination of folic acid therapy the above routine was carried out for a minimum of 2 weeks if possible. The follow-up studies of Group B patients (the leukopenias) differed only in that daily white blood counts and blood smear examinations were done instead of daily reticulocyte counts. All blood counts were done with Bureau of Standards equipment. The diagnoses as listed in Tables 2 and 3 were well established on the basis of both clinical and laboratory evidence and the diagnosis of malignant disease was in each case supported by examination of suitable pathologic material.

Results. The results of administration of folic acid are summarized in Tables 2 and 3.

1. Five cases of pernicious anemia in relapse have been treated with folic acid (see Cases 1, 2, 3, 4, 5, Table 2, Chart 1). All of these cases obtained a satisfactory

TABLE 1.—FOLIC ACID GROUP OF SUBSTANCES*

I Investigators	II Date	III Name	IV Biologic activity
1. Wills and Bilimoria[25]	1932–1935	Vitamin B	Cures or prevents nutritional cytopenia in monkeys
2. Day and Langston[5,6]	1935–1938	Vitamin M	Same as above
3. Stokstad and Manning[21]	1938	Vitamin B	Cures chick anemia
Hogan and Parrott[11,12]	1939		
4. Snell and Peterson[18]	1940	Norite eluate factor	Growth factor for *L. casei* and *S. lactis*
5. Hutchings, Stokstad, Bohonos and Slobodkin[13]	1943–1944	*L. casei* factor	Growth factor for *L. casei* and *S. lactis*, action similar to: vitamin B, norite eluate factor, and vitamin M
6. Mitchell, Snell and Williams[17]	1941	Folic acid	A potent hemopoietic factor
7. Angier *et al.*[1]	1945–1946	Synthetic folic acid and (formula)	Similar to "natural" folic acid

* This is not intended to be a complete survey of all the substances and of all the investigators concerned with this substance.

Early in 1945 synthetic folic acid* became available in large enough quantity to enable us to treat a number of leukopenic and anemic states.

Method. The patients were divided into 2 groups, A and B. Group A patients were those in whom the primary consideration

hematologic remission in their disease following the administration of folic acid. On the average, the reticulocyte response began on the 4th to the 5th day following institution of treatment, reached a peak on the 7th or 8th day and slowly fell to pre-treatment levels within 2 weeks.

* Courtesy of Dr. S. M. Hardy of Lederle Laboratories, Inc., New York, New York.

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TABLE 2

TABLE 2.—GROUP A PATIENTS IN WHOM THE MAIN CONSIDERATION WAS THE RESPONSE OF THE RED BLOOD CELL COUNT AND HEMOGLOBIN TO THE ADMINISTRATION OF FOLIC ACID

Case	Diagnosis	Admission R.B.C. (mill. per c.mm.)	Discharge R.B.C. (mill. per c.mm.)	Admission Hgb. (gm. per 100 cc.)	Discharge Hgb. (gm. per 100 cc.)	Peak retic. count (%)	Expected peak retic. count with intramuscular liver (%)	Amount of folic acid (mg. per day)	Route of administration	Total amount of folic acid (mg.)	Effect
1	Pernicious anemia	1.35	3.94	5.4	11.7	28.2	31.2	15	Intramuscularly	240	Excellent
2	Pernicious anemia	0.71	2.98	2.7	11.5	16.4	49.0	50	Orally	750	Excellent
3	Pernicious anemia	0.874	2.95	2.6	9.7	21.8	43.6	50	Orally	750	Excellent
4	Pernicious anemia	1.15	3.00	5.5	10.1	24.2	36.0	50	Orally	750	Excellent
5	Pernicious anemia	1.66	2.47	8.0	11.7	15.4	24.8	10	Orally	180	Excellent
6	Sprue	3.50	5.01	13.1	14.7	2.3	..	40	Intramuscularly	1150	Good
7	Macrocytic anemia, unknown origin	2.63	2.10	9.4	7.3	4.0	..	45	Orally	495	Poor
8	Macrocytic anemia, unknown origin	2.30	3.40	10.3	12.0	4.9	..	50	Orally	900	Questionable
9	Macrocytic anemia, unknown origin	2.37	2.20	8.8	8.5	3.4	..	10	Orally	120	Poor
10	Anemia of renal insufficiency	2.37	3.15	7.5	8.4	2.1	..	50	Orally	730	Poor
11	Anemia of renal insufficiency	1.80	1.82	6.3	6.5	6.0	..	50	Orally	750	Poor
12	Anemia of renal insufficiency	2.80	2.50	9.7	8.5	3.0	..	50	Orally	750	Poor
13	Anemia of renal insufficiency	2.69	2.73	7.3	8.2	2.5	..	20	Orally	320	Poor

TABLE 3.—GROUP B PATIENTS IN WHOM THE MAIN CONSIDERATION WAS THE RESPONSE OF THE WHITE BLOOD CELL COUNT TO THE ADMINISTRATION OF FOLIC ACID

Case	Diagnosis	Admission W.B.C.	Discharge W.B.C.	Amount of folic acid (mg. per day)	Route of administration	Total amount of folic acid (mg.)	Duration of treatment (days)	Effect
14	Addison's disease	4880	3,500	15	Orally	50	10	Poor
15	Felty's syndrome	1790	1,350	50	Orally	700	14	Poor
16	Hyperthyroidism, thiouracil agranulocytosis	1080	8,500	50	Orally	550	11	Questionable
17	Idiopathic leukopenia	3600	4,400	50	Orally	500	10	Poor
18	Idiopathic leukopenia	1400	1,500	5–15	Orally	125	14	Poor
19	Idiopathic leukopenia	3400	2,700	5–15	Subcutaneous & Orally	270	42	Questionable
20	Idiopathic leukopenia	4100	2,100	50	Orally	500	10	Poor
21	Idiopathic leukopenia	1800	2,500	10	Orally	80	8	Poor
22	Idiopathic leukopenia	2800	2,800	10	Orally	100	10	Poor
23	Idiopathic leukopenia	2500	2,050	20	Intramuscularly	280	14	Poor
24	Irradiation leukopenia, lymphatic leukemia	2600	14,950	5–15	Orally	175	15	See Chart 6
25	Acute leukemia	2500	2,700	20	Subcutaneous & Orally	460	23	Poor
26	Irradiation leukopenia, lymphosarcoma	2700	2,000	20	Orally	200	10	Poor
27	Irradiation leukopenia, Hodgkin's disease	1800	2,800	50	Orally	700	14	Poor
28	Irradiation leukopenia, malignant lymphoma	2200	1,800	50	Orally	500	10	Poor
29	Irradiation leukopenia, cancer of breast	3500	2,500	10	Orally	60	6	Poor
30	Irradiation leukopenia, cancer of thyroid	4800	4,800	15	Orally	150	10	Poor
31	Irradiation leukopenia, cancer of breast	4300	3,300	15	Orally	150	10	Poor
32	Irradiation leukopenia, lymphosarcoma	3400	4,100	50	Orally	500	10	Poor
33	Irradiation leukopenia, cancer of cervix	3300	2,800	15	Orally	150	10	Poor
34	Irradiation leukopenia, lymphosarcoma	3400	2,400	50	Orally	500	10	Poor

Within 6 to 8 weeks the red blood cell count had increased on the average to 292% and the hemoglobin to 269% of the values prior to giving folic acid. No direct counts were made on the number of blood platelets, although from observations made from examination of blood smears, these elements were also noted to increase in number. At the end of 2 to 4 weeks of folic acid therapy, 4 of the 5 cases of pernicious anemia were given parenteral liver extract. No subsequent reticulocyte response was noted in any of these cases.

preparation, it became necessary to give the patient whole blood transfusions and parenteral medication (see Chart 2).

3. Case 6 is the only patient with sprue which we have had the opportunity to treat. As can be seen in Chart 3, the red blood cell count rose from 3.5 million per c.mm. to 5.01 million per c.mm., and the hemoglobin rose from 13.1 gm. % to 14.7 gm. % in $7\frac{1}{2}$ weeks. No reticulocyte response was noted. On the other hand, the clinical improvement was striking for the patient gained strength and weight and his diarrhea subsided.

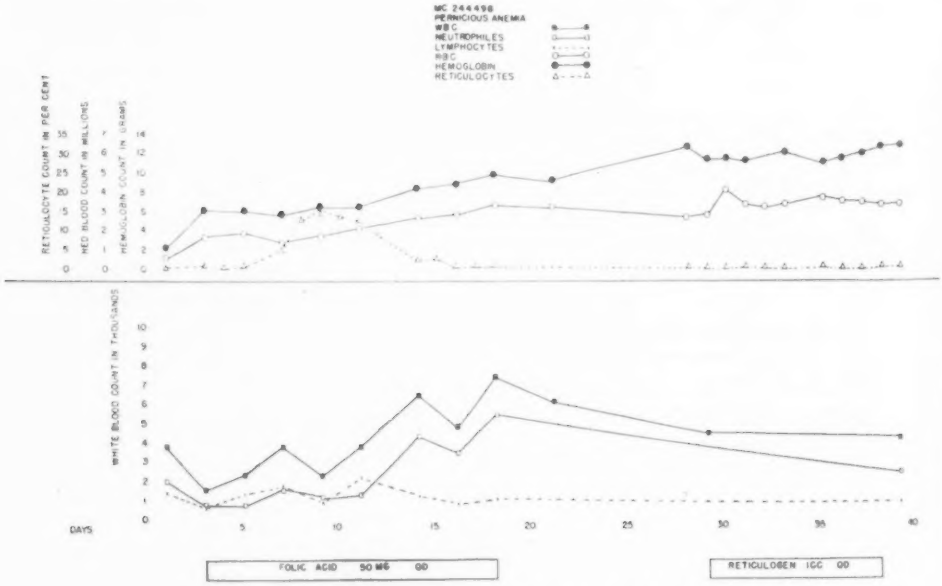

CHART 1.—Response to folic acid in a case of pernicious anemia.

2. The effect of combined preparation of folic acid and iron on a patient with pernicious anemia is represented in Case 1. This preparation was later found to be low in folic acid activity. On the 3rd day of treatment with this combined preparation, the patient developed diarrhea and absorbed little, if any, folic acid, for there was no reticulocyte response and the bone marrow remained megaloblastic in type. The clinical condition became rapidly worse and finally on the 5th day following institution of the combined folic acid-iron

4. In 3 cases of macrocytic anemia of undetermined origin no effect was produced by administration of folic acid (see Cases 7, 8 and 9, Table 2).

5. No beneficial effects were noted on the anemia of 4 cases of renal insufficiency treated with folic acid (see Cases 10, 11, 12, 13, Table 2, Chart 4).

6. The administration of folic acid produced no effect on the leukopenia associated with Addison's disease (1 case) (see Case 14, Table 3).

7. In 1 case of Felty's syndrome no

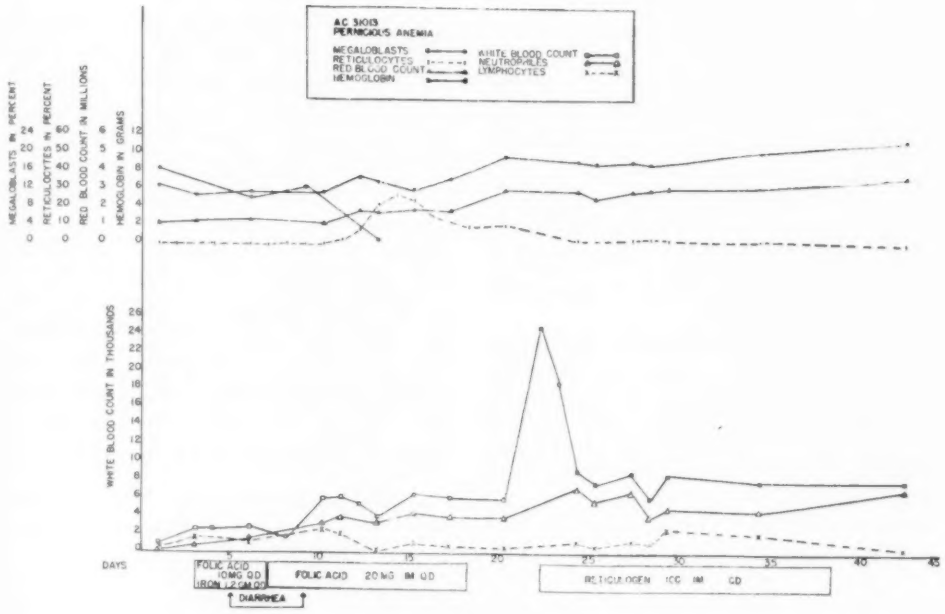

CHART 2.—Delayed response to combined folic acid iron preparation, perhaps due to the interference of iron with the absorption of folic acid.

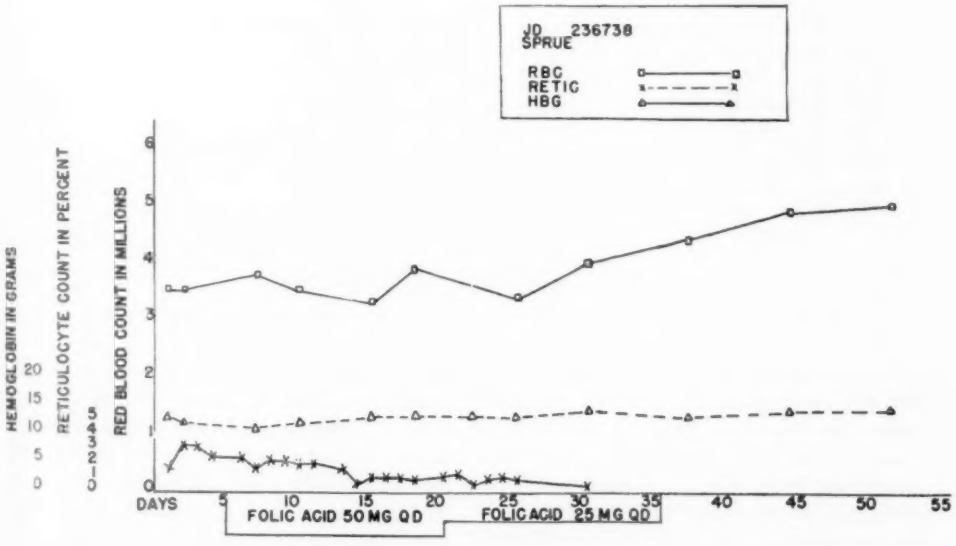

CHART 3.—Response to folic acid in a case of sprue.

effect of folic acid was detected on either the white or red blood cell count (see Case 15, Table 3).

8. In 1 case of agranulocytosis resulting from thiouracil administration for hyperthyroidism, an increase of the white blood cell count was observed 7 days following the oral administration of 50 mg. of folic acid. It should be emphasized, however, that thiouracil had been discontinued at the same time that folic acid was begun (see Case 16, Table 3).

9. Seven cases of idiopathic leukopenia have been studied. These cases were not suffering from the lack of any known nutritional factors, had not been exposed to any material known to be toxic to the following the administration of oral folic acid. Despite these responses this patient ultimately died and the leukocytosis, although apparently associated with folic acid administration was not permanent. The final counts lapsed to their former levels despite continuation of treatment.

10. Two cases of leukemia have been given folic acid. Case 24, a patient with chronic lymphatic leukemia, was given folic acid because of leukopenia induced by Roentgen radiation. At the start of folic acid treatment his count was 2600. Four days after receiving folic acid (total amount 20 mg.) his white blood cell count had risen to 8000 per c.mm. A second course of folic acid given 1 month later

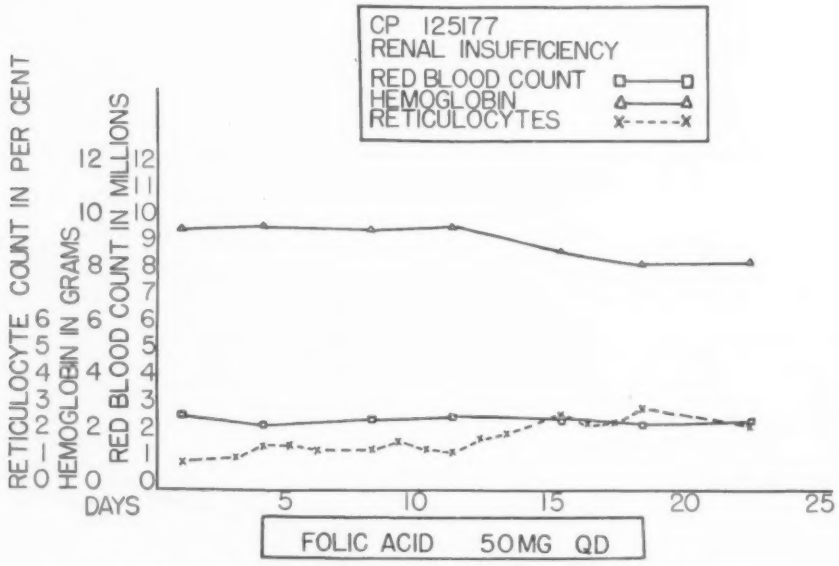

CHART 4.—Failure to respond to folic acid in a case of anemia of renal insufficiency.

bone marrow and on physical examination did not have palpable spleens. Despite numerous diagnostic tests, no satisfactory diagnosis could be established. As can be seen from Table 3 no apparent beneficial effect can be attributed to the administration of folic acid in these cases with the possible exception of Case 19, Chart 5. This child showed a slight, but detectable leukocytic response on at least 2 occasions had a slight effect on the white blood cell count (see Case 24, Table 3, Chart 6).

Case 25 was a patient with acute myeloblastic leukemia who received 20 mg. of folic acid daily over a period of 23 days. Although there was a transient rise in the hemoglobin and red blood cell count following administration of this drug, the patient succumbed 4 months after the onset of the disease.

11. Nine cases of leukopenia resulting from Roentgen radiation because of various types of malignant disease have been studied. In none of these cases was any effect noted on the level of the white blood cell count following folic acid administration. Chart 7 is representative of this negative effect (see also Cases 26 to 34, Table 3).

Discussion. 1. In our series of cases the best results of folic acid therapy were obtained in cases of pernicious anemia in relapse (see Chart 1). In general, the hematologic response parallelled, but was not identical with, the response obtained in liver-treated cases. These differences have been pointed out by others, Amill and Wright,[3] and Frommeyer and Spies,[9] and Meyer[15] who have shown that the reticulocyte response following folic acid administration differs from what might be expected following the administration of active liver principle. Chart 8 shows this difference very clearly. In 5 cases of pernicious anemia treated with folic acid, the mean reticulocyte curve was not as high as was obtained in a comparable group of cases of pernicious anemia treated with potent liver extract. The chart also shows that in the folic acid treated group the reticulocytes tended to remain elevated over a longer period of time.

Recently Frommeyer and Spies[9] and Heinle and Welch[10] have observed that folic acid may not control the neurologic manifestations of pernicious anemia. In fact neurologic relapses have developed during folic acid administration. Although we have had no experience with the prolonged treatment of pernicious anemia

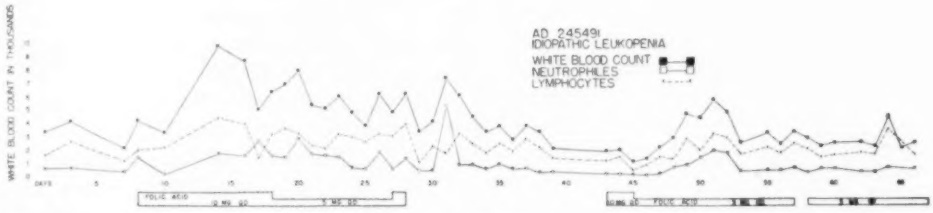

CHART 5.—Questionable temporary effect on the white blood count in a case of idiopathic leukopenia in an infant.

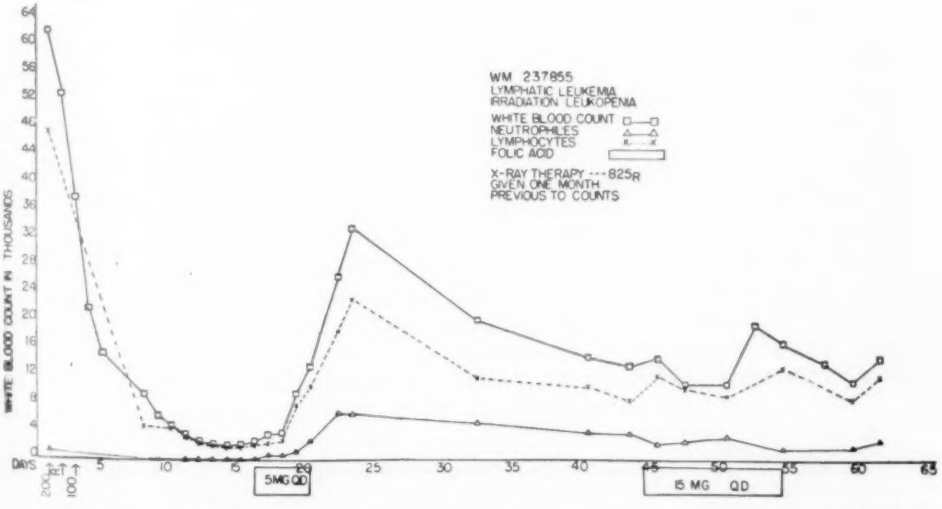

CHART 6.—The effect of folic acid upon a case of chronic lymphatic leukemia in which leukopenia resulted from Roentgen treatment.

with folic acid it has been observed by us that the paresthesias so characteristic of this disease show little tendency to clear following administration of this drug (see Case 5). The answer to this interesting problem will have to await further study.

2. In this group of cases of pernicious anemia, Case 1 was of particular interest to us. This patient received a combined folic acid-iron preparation which was later proved to be low in folic acid activity. The iron caused diarrhea and thereby might have impaired the absorption of folic acid given by mouth. This fact was supported by the observations that no reticulocyte increase was detected and that the bone marrow remained megaloblastic in type.

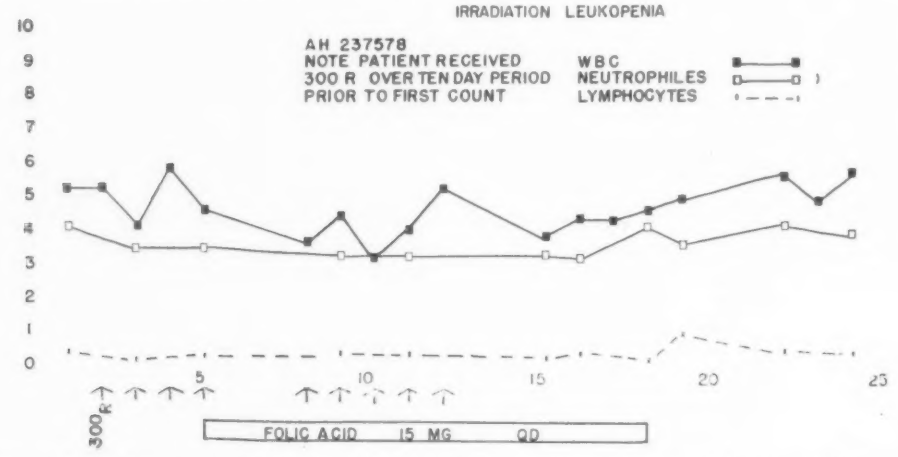

CHART 7.—Failure to respond to folic acid in a case of metastatic carcinoma of the thyroid given Roentgen treatment.

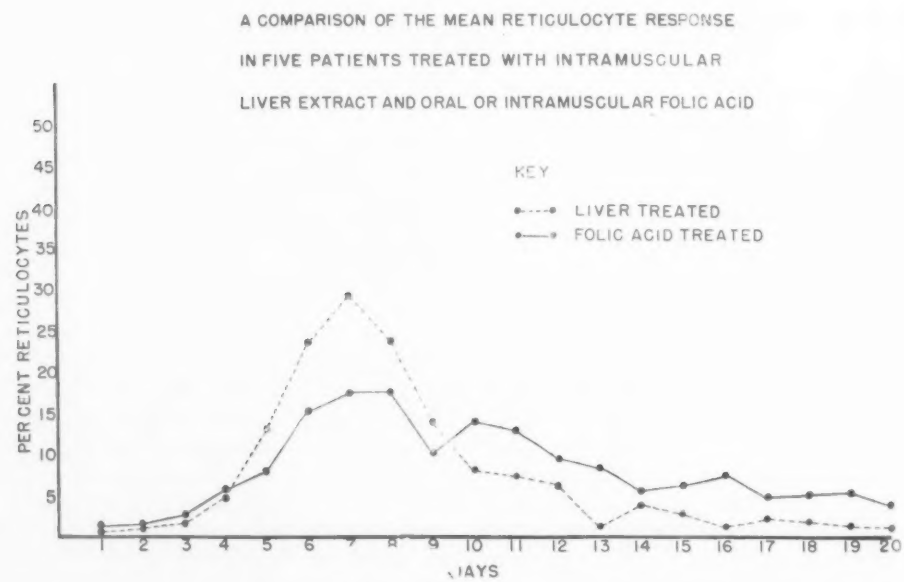

CHART 8.—Self-explanatory.

It is true that in some cases of pernicious anemia the iron stores may be depleted, but since this is not the fundamental defect in this disease it seems unwise to jeopardize the absorption of folic acid by the use of a combination of this drug with iron (see Chart 2).

3. The favorable effect of folic acid upon sprue has been reported by Darby and Jones,[4] and Spies, Menendez *et al.*[19] who have shown marked hematologic and clinical improvement in patients who were treated for this disorder. For experimental purposes their patients were placed on diets low in protein and poor in vitamin B substances and were then given folic acid. Prompt clinical and hematologic improvement occurred. Their patients gained strength and weight, exhibited a pronounced reticulocyte response and experienced a cessation of their distressing bowel complaints. Except for failure to produce a reticulocyte response, our single case showed a similar tendency to improve following folic acid administration. It can be seen in Chart 3 that the patient's red blood cell count rose slowly. Perhaps the reason why a more striking hematologic improvement was not noted was because the patient was not profoundly anemic upon admission. However, the clinical improvement was quite satisfactory.

4. The mere existence of a macrocytic anemia does not imply that there will be a beneficial response to folic acid. For example, the 3 cases of macrocytic anemia of undetermined origin (Cases 7, 8 and 9) showed no response to folic acid administration. No elevation of the reticulocyte count or the red blood cell count was noted in these cases. In our experience, as in the experience of others,[21,22] the seeming prerequisite for a beneficial response to folic acid on the part of the red blood cell picture is a megaloblastic arrest as seen in the bone marrow. This may not be true in all cases but for the present at least this observation seems to be correct.

5. Numerous theories have been propounded as to the mechanisms of the production of anemia in chronic nephritis. Hematuria, bleeding into the tissues, hemolysis, loss of albumin, dietary deficiency in protein, direct toxic action on the bone marrow, insufficient blood regeneration, have all been implicated. Those who have studied the bone marrow in this type of anemia seem to agree that ultimately there is a selective erythroid hypoplasia which in some manner is associated with the azotemia. It is generally accepted that the severity of the erythroid hypoplasia and anemia is correlated with the degree and duration of the azotemia. In our cases early in the course of chronic nephritis the bone marrow may be normoblastic in type but subsequently there is an exhaustion of the erythroid elements and a tendency towards hypoplasia. It is interesting to note in our series of cases there was a slight but persistent reticulocytosis prior to the administration of folic acid. In none of the 4 cases in which this drug was used was there any hematologic response. We have no good explanation for the anemia of renal insufficiency or for the reticulocytosis which we have observed in this condition. We believe that a more extensive search should be made for a hemolytic factor which may play some rôle in this type of anemia.

6. It has been stated by Albright *et al.*[7] that patients with Addison's disease exhibit a relative neutropenia and lymphocytosis in comparison with normal figures. Accordingly, it was thought to be of interest to study the effect of folic acid in this condition. No effect was noted on either the total white blood cell count or the differential formula in the 1 case which has been treated. Although the fundamental hemopoietic defect which may cause leukopenia in Addison's disease is unknown, we would expect that folic acid would not correct the neutropenia which sometimes occurs in this condition.

7. Little is known concerning the mechanism of leukopenia in Felty's syndrome. It is known that splenectomy does not produce lasting benefit in these

cases so that so-called "hypersplenism" is probably not an important factor. In the 1 case which was treated, folic acid had no effect on either the anemia or the leukopenia.

8. One case of agranulocytosis resulting from administration of thiouracil was studied. The apparent effect of folic acid in bringing about an elevation of the total white blood cell count in this case, we do not wish to attribute to folic acid. It was probably the result of removal of the patient from thiouracil treatment. The evidence is good at the present time that folic acid is neither of benefit as a prophylactic or therapeutic agent for the agranulocytosis produced by thiouracil.

9. Seven cases of "idiopathic" leukopenia have been treated. In 6 cases the results of treatment were entirely negative. There was no alteration of the total white blood cell count or the differential formula. In Case 19 (see Chart 5) there seemed to be a response to folic acid as the total white blood cell count and the number of granulocytes increased in number on 2 separate occasions. However, this effect was transitory and the patient ultimately succumbed despite continuation of folic acid medication. The poor results obtained in this group are consistent with the findings of Doan,[8] and Spies[20] who have noted little or no effect of folic acid in cases of "idiopathic" leukopenia treated with folic acid.

10. We have treated 2 cases of leukemia with folic acid. As noted under Results, Case 24 was a patient with chronic lymphatic leukemia who was given folic acid because of a leukopenia induced by Roentgen radiation. The white blood cell count returned promptly to high levels (see Chart 6). It cannot be said whether this represented a true response to folic acid or whether it was mere coincidence.

The second case of leukemia which has been studied—a case of acute myeloblastic leukemia (Case 25)—was given folic acid in a leukopenic phase of the disease. Despite an apparent remission characterized by a feeling of well-being and an increase in the red blood cell count and hemoglobin values, this patient succumbed 4 months after the recognition of his disease. This poor result is typical of the findings of others[20,26] who have obtained similar observations in the folic acid treatment of leukemia. However, as suggested by Case 24, there may be some rationale in giving folic acid to those cases of leukemia which develop leukopenia in the course of Roentgen ray therapy. This, of course, cannot be established from the findings in 1 case.

11. Many investigators[12,14,24,25] have shown the importance of folic acid in maintaining the normal white blood cell count. Considerable importance has also been attached to the study of the living organisms' ability to respond to folic acid when exposed to certain conditions of stress. One of the most important of these studies has been the surprising observations of Watson, Sebrell, McKelvey and Daft[22] who noted the correction of leukopenia induced by Roentgen radiation following the administration of folic acid. They reported favorable results in 7 out of 8 cases so treated, but state their results were not conclusive and that further investigation was indicated. Nine cases of leukopenia of this type have been treated. In none of these cases have we obtained a correction of the leukopenia by the use of folic acid. It is difficult to understand how folic acid would be of use in correcting this type of leukopenia, but it must be admitted that in a certain few cases there may be a nutritional factor which is in part responsible for the low white blood cell counts. In these certain few cases folic acid may be of some benefit.

Summary. Results of a study of 34 cases of varying types of blood dyscrasias treated with folic acid show that: 1. Folic acid will correct the abnormal blood findings in cases of pernicious anemia in relapse.

2. It is likely that the neurologic manifestations of pernicious anemia are not

corrected by folic acid.

3. In pernicious anemia the reticulocyte response in liver treated patients differs from that obtained in folic acid treated patients.

4. It is probably unwise to combine folic acid and iron in the treatment of pernicious anemia.

5. Sprue responded favorably to folic acid administration (1 case).

6. All macrocytic anemias do not respond to the administration of folic acid. In those cases in which there is a megaloblastic arrest in the bone marrow, favorable results can be anticipated.

7. Folic acid did not affect the anemia associated with chronic nephritis.

8. The administration of folic acid did not alter the leukopenia associated with either Addison's disease or Felty's syndrome (1 case each).

9. The apparent effect of folic acid upon the level of granulocytes is 1 case of thiouracil agranulocytosis was probably coincidence for others have reported no effect.

10. Folic acid had no effect on the leukopenia of idiopathic states.

11. No beneficial effect was noted in 2 cases of leukemia treated with folic acid.

12. Folic acid did not correct the leukopenia induced by Roentgen radiation.

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PHARMACODYNAMIC PROPERTIES OF PENICILLIN PREPARATIONS WITH PROLONGED ACTION

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We suggested recently[6] a penicillin mixture for clinical use which has the advantage of prolonged action, high fluidity, and readiness for injection with an ordinary syringe. The standard preparation recommended for human use consists of a suspension of 300,000 u. crystalline potassium penicillin in 1 cc. vegetable oil containing 0.3 mg. epinephrine. This is a milky suspension, available in vials and ampules, which can be injected with a 20 to 21 gauge needle.

A clinical evaluation of the efficiency of this suspension in cases of male gonorrhea, conducted by Cohn and Kornblith,[3] confirmed the advantages claimed. Investigations on the activity of this penicillin composition in other infections are in progress: one may anticipate that the clinical results will be parallel to those of Cohn and Kornblith, *i. e.*, increased activity, determined by the repository effect.

The present paper contains a number of observations gained in the course of our study; some of these have a general interest concerning the relationships (*a*) between dosage and penicillin levels, rate of absorption, dosage and animal species, size of the individual, etc.; (*b*) between blood and tissue levels; and (*c*) between systemic and local effects of the vasoconstrictor.

Material and Methods. In dog, rabbit and man, 1 or 2 cc. of the material was injected intramuscularly (if not otherwise stated) into the buttocks. In the rat 0.1 cc. was consistently injected intramuscularly. Blood samples were withdrawn at various intervals after treatment and their penicillin content determined by the serial dilution method,[11] using rabbit blood as indicator for the growth of *Strep. hemolyticus*, Type A, Group 3. The minimal penicillin concentration which inhibited the growth of this microorganism varied between 0.015 and 0.03 u. per cc., which is the usual sensitivity of the method. The blood was withdrawn from the jugular vein of dogs and rabbits, from the subaxillary artery of the anesthetized rat, and from the cubital vein of man. The organs of rats were removed after the animal was thoroughly bled; their penicillin content was determined with the dilution method on the tissue bray, ground with sand in a mortar and diluted with twice its weight in 1% phosphate buffer pH 6.

Results. The following observations are based on approximately 200 experiments on dogs, 120 on man, 50 on rabbits and 400 on rats.

1. Influence of the Vehicle on the Blood Level. We determined, in the first place, the influence of the vehicle in which the penicillin is injected on the blood levels. Representative data for the following media are summarized in Figure 1: (*a*) saline, (*b*) peanut oil, (*c*) water/oil emulsion similar to that recommended by Freund and Thomson:[8] (peanut oil + 25% of a cholesterol ester preparation (Falba or Amerchol) emulsified by the addition of 25 to 50% saline), (*d*) peanut oil + 4.8% beeswax, and (*e*) ethyl oleate + 4% beeswax.

It appears that with the penicillin dosages used in clinical practice (200,000 to 300,000 units) oil alone and the water/oil emulsion give no prolongation of the blood level compared to the administration of aqueous solutions of equal dosage. This observation is in agreement with the report of Harris, Wilcox and Finland.[9] Similarly, aqueous gelatin, up to 50%, does not induce any prolongation of the penicillin level. The peanut oil beeswax mixture, recommended by Romansky[12] exerts, as known, a great retaining power which is entirely due to its beeswax content.

Two cc. of 4% beeswax in ethyloleate containing 250,000 u. Ca-penicillin (English preparation) gave shorter durations than 300,000 u. in 1 cc. of 4.8% in peanut oil. The shorter duration of the English preparation probably depends on the relative concentration of penicillin per cc. as the experiments of Ungar[14] suggest rather than on the dosage difference.

2. Influence of the Vasoconstrictor Dosage on the Blood Level. After having established the influence which the character of the vehicle exerts on the absorption speed of intramuscularly injected penicillin, the effect of vasoconstrictors incorporated into the various media was studied.

In the early part of the investigation, we noticed that extremely small doses of epinephrine incorporated in the emulsion, or in the oil, induced a very definite prolongation of the blood level. Since a mixture of penicillin and epinephrine can be prepared as a stable oily suspension, we determined on experimental animals the quantitative relation between the duration of penicillin blood level and the epinephrine content. (Fig. 2.) A prolonging action of 0.01 mg. epinephrine on the duration of the blood level was already noticeable; this action increases with increasing epinephrine dosage, up to a "ceiling" dosage (0.2 to 0.3 mg.), where a further increase of epinephrine is without effect on the duration. The ceiling in the prolonging effect of the vasoconstrictor might depend on a *maximal* ischemic reaction of the surrounding tissue.

In man we compared the effect of 0.25 mg. and 1 mg. epinephrine given subcutaneously with 300,000 units penicillin in 1 cc. oil. The differences found in duration were in the range of the individual variations. Thus it seems that this ceiling dose for the local effect of epinephrine is the same in man as in dog.

3. Influence of the Penicillin Dosage (in Oil With Vasoconstrictor). The blood level curves seem to indicate that following penicillin administration in oil containing vasoconstrictor the duration of the blood level increases proportionately up to only a certain penicillin dosage. This ceiling dosage is approximately 200,000 u. for the dog and 300,000 u. for man, which in the 2 species gives a *1 day* level.

In man, by increasing the intramuscular dose from 200,000 to 300,000 u., the length of the blood level is increased by 5 to 9 hours, while the increase of duration is only from 1½ to 3 hours when the 300,000 u. dose is brought to 600,000 u. This increased dosage induces, however, without substantially influencing the duration, blood penicillin curves running at higher levels.

The blood levels following the administration of penicillin in oil with vasoconstrictor are quite high for a prolonged period (10 to 15 hours), after which the progressive descent of the blood curve starts. Figure 3 shows the great increase of the blood levels in man during the first 15 hours following the administration of penicillin in oil + vasoconstrictor as compared to saline.

Similar are the observations on dogs. By increasing the 200,000 u. dosage to 300,000 u. the average duration increases from 18½ to 22 hours; by increasing it to 400,000 u. the duration reaches up to 24 hours.

In dog, as in man, by the use of an oily vehicle with vasoconstrictor high initial blood levels were maintained for long periods, generally for over 10 hours. The following examples illustrate the extent of this high initial "plateau:"

Dog No. 2039—weight 9.6 kg.

Injected with 200,000 u. potassium penicillin in 1 cc. oil and 0.05 mg. epinephrine:

Hours after injection	Serum levels (units/cc.)
2½	4.0
4½	12.0
6½	4.0
16	0.5

Dog No. 2056—weight 8 kg.

Injected with 300,000 u. potassium penicillin in 1 cc. oil and 0.2 mg. epinephrine:

Hours after injection	Serum levels (units/cc.)
1	9 0
3	20 0
7	8 0
17	1 0

The same dog, No. 2056, injected with a higher dose, *i. e.*, 400,000 u. crystalline potassium penicillin in saline, has, in contrast, the following concentration:

Hours after injection	Serum levels (units/cc.)
1	9 0
3	9 0
7	0 03
8½	0 0

Two other important facts are visible from these examples, namely, that in the dog at least, following the injection of penicillin in oil with vasoconstrictor, the blood concentration reaches the same height as that obtained with penicillin injected in saline, though the appearance of the peak is delayed. In the examples presented, the peak effect is reached 3 to 4½ hours after injection of the oily suspension with vasoconstrictor. This value represents the average recorded in our various experiments: minimum 2 hours; maximum 7 hours.

In our animal experiments we do not find any impressive differences between various penicillin salts (crystalline potassium and sodium penicillin; amorphous calcium penicillin) when suspended in oil epinephrine medium in regard to the duration of the blood level. However, a somewhat shorter duration following intramuscular injection of calcium penicillin into man is found by Dr. Queally in comparison to that seen with the crystalline potassium salt. (The durations with Ca-penicillin in man are similar to those obtained with commercial preparations of Ca-penicillin in peanut oil containing 4.8% beeswax.) This finding is in agreement with various reports concerning the shorter blood level following administration of the amorphous calcium salt of penicillin compared to the crystalline material administered in this mixture.[5] The particle size of the crystalline material has no influence on the duration in dogs injected intramuscularly with 300,000 u. This is at variance with the results reported for the liquid POB mixture, and similar to the findings with the solid mixture.[5]

4. Blood Level in Different Species. The duration of the demonstrable blood penicillin level is approximately the same in man and dog, if equal doses (300,000 to 600,000 u.) of potassium penicillin in oil epinephrine are given. Accordingly, the minimal dose required to obtain a 1 day duration calculated *on the basis of body weight* is inversely proportional to the size of the species. For the rat this dose is 100,000 u. per kg.; for the rabbit about 30,000 u. per kg.; for the dog 25,000 u. per kg.; for man 4000 to 5000 u. per kg.

Generally speaking, it is not the *relative* but the *absolute* dose injected which determines the prolongation of the penicillin effect. This is particularly apparent in observing the duration following the injection into dogs of doses used in human therapy.

The same total amount of penicillin absorbed during an approximately equal time period—considering a fairly similar rate of elimination—results in higher blood concentrations in the species of smaller size. This statement is based on the figures of Table 1.

The individual variations are as wide in experimental animals as in man. For instance, in dogs treated with 300,000 u. (in 1 cc. oil + 0.2 to 0.4 mg. epinephrine) the shortest blood level is 16 hours duration, the longest 26 hours. In man, treated subcutaneously with 600,000 u., the shortest duration is 18 hours, the highest so far recorded over 25 hours.

5. Duration of Penicillin in Tissues. The urines of the subjects treated with penicillin in oil with vasoconstrictor contian relatively high amounts of penicillin after the drug ceases to be detectable in the blood. The following example is characteristic: 3 male subjects treated Tuesday 10 A.M., with 300,000 u. in 1 cc. oil containing 0.3 mg.

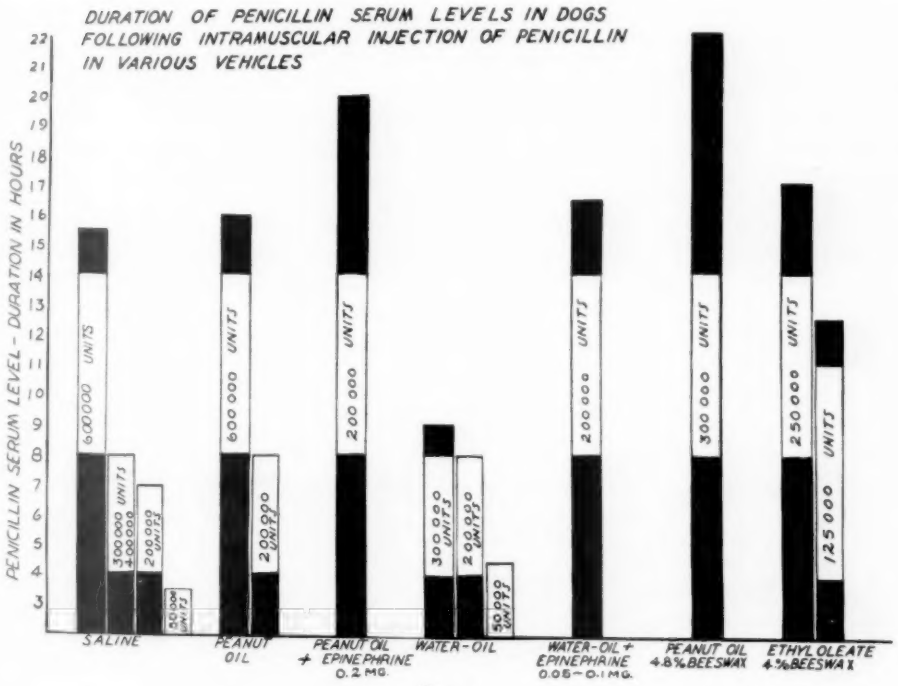
DURATION OF PENICILLIN SERUM LEVELS IN DOGS FOLLOWING INTRAMUSCULAR INJECTION OF PENICILLIN IN VARIOUS VEHICLES
PENICILLIN SERUM LEVEL - DURATION IN HOURS
600000 UNITS
300000 UNITS + 100000
200000 UNITS
50000 UNITS
600000 UNITS
200000 UNITS
200000 UNITS
300000 UNITS
200000 UNITS
50000 UNITS
200000 UNITS
300000 UNITS
250000 UNITS
125000 UNITS
SALINE
PEANUT OIL
PEANUT OIL + EPINEPHRINE 0.2 MG.
WATER-OIL
WATER-OIL + EPINEPHRINE 0.05-0.1 MG.
PEANUT OIL 4.8% BEESWAX
ETHYL OLEATE 4% BEESWAX

FIG. 1

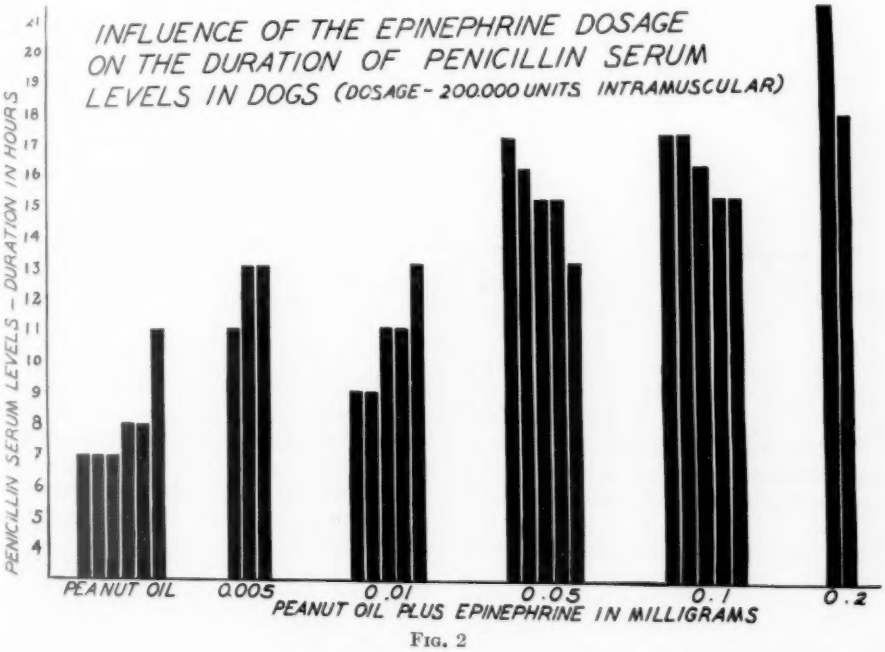
INFLUENCE OF THE EPINEPHRINE DOSAGE ON THE DURATION OF PENICILLIN SERUM LEVELS IN DOGS (DOSAGE - 200.000 UNITS INTRAMUSCULAR)
PENICILLIN SERUM LEVELS - DURATION IN HOURS
PEANUT OIL
0.005
0.01
0.05
0.1
0.2
PEANUT OIL PLUS EPINEPHRINE IN MILLIGRAMS

FIG. 2

epinephrine; 7 hours after injection: blood level 0.63, urine 375 to 600 u.; Wednesday NOON: blood samples negative; urine samples: 0.5 to 1.8 u. per cc.; Thursday NOON: 0.1 u. per cc. in urine. This delayed elimination of penicillin in the urine seems to be a general characteristic of repository penicillin administration, since similar observations were reported for the peanut oil beeswax mixture by Romansky.[12,13]

The presence of penicillin in the urine after the blood became negative led us to investigate its duration in the tissues. Experiments on rats reveal that the duration of the penicillin level is much more prolonged in the liver and kidney than in the blood. (See Fig. 4.)

oil containing 0.06 mg. epinephrine given 3 hours before injection protected the majority of mice. This is a high degree of prophylactic effect compared to that of an aqueous solution, which gives no protection at all in doses of 2000 u. per 20 gm.

A certain improvement in the protective effect was noticed also using peanut oil suspensions of penicillin. To obtain this effect, however, the doses of penicillin required are 2 times higher than in the presence of a vasoconstrictor.

(*b*) *Experimental Rabbit Syphilis.* Rabbits with acute testicular lesions, infected as described by Ercoli and Lafferty,[7] were treated intramuscularly with various doses of penicillin in oil with epinephrine.

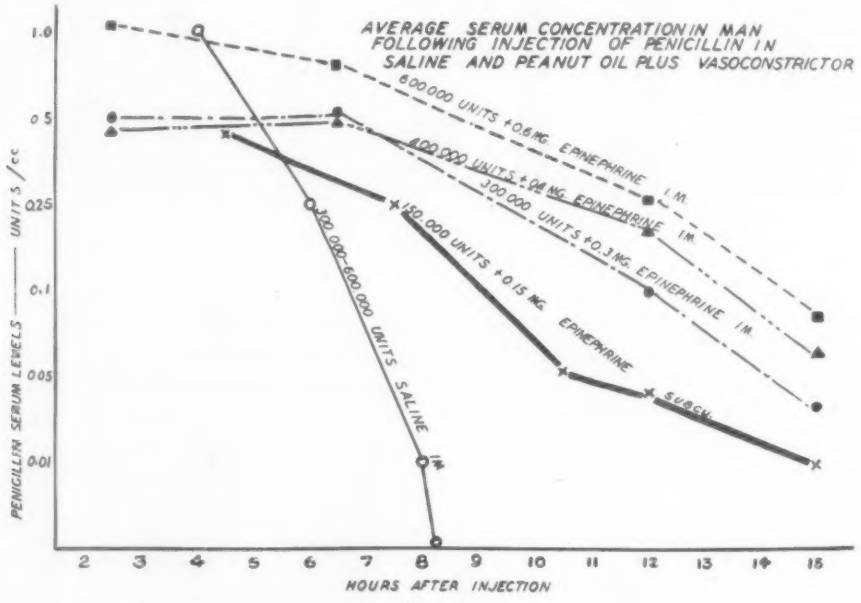
AVERAGE SERUM CONCENTRATION IN MAN FOLLOWING INJECTION OF PENICILLIN IN SALINE AND PEANUT OIL PLUS VASOCONSTRICTOR
600,000 UNITS +0.6 MG. EPINEPHRINE I.M.
400,000 UNITS +0.4 MG. EPINEPHRINE I.M.
300,000 UNITS +0.3 MG. EPINEPHRINE I.M.
150,000 UNITS +0.15 MG. EPINEPHRINE S.U.&C.M.
600,000 UNITS SALINE I.M.
PENICILLIN SERUM LEVELS —— UNITS/cc
1.0
0.5
0.25
0.1
0.05
0.01
2 3 4 5 6 7 8 9 10 11 12 13 14 15
HOURS AFTER INJECTION

FIG. 3

6. CHEMOTHERAPEUTIC ACTIVITY IN EXPERIMENTAL INFECTIONS. (*a*) *Pneumococcus Type II.* The duration of the *prophylactic* effect of various doses of penicillin in oil + vasoconstrictor was compared with that following administration in saline or in oil alone.

Mice were infected intraperitoneally with 1000 lethal doses of pneumococcus 6302, as described by Buck and Schnitzer,[1] at various intervals after subcutaneous treatment. Doses of 500 u. per 20 gm. injected in 0.5 cc.

The smallest dose used, 1 single injection with 10,000 to 25,000 u. per kg., induced rapid disappearance (6 to 12 hours) of the spirochetes and regression of the syphilitic lesion (4 to 8 days) in all 6 rabbits treated. For the quantitative evaluation of curative activity we must await the results of the lymph node transfer.

7. PHARMACOLOGIC EVALUATION OF VASOPRESSOR ACTION. As known,[10] the oily vehicle interferes with the intensity of the systemic action of epinephrine. We re-

ported[6] the absence of vasopressor activity in man of penicillin preparations containing 0.6 to 0.8 mg. epinephrine, *i. e.*, twice the dose contained in the standard preparation. A large number of tests on anesthetized cats, carried out by Dr. R. J. Schachter, with the usual pharmacologic methods, showed that the intramuscular injection of 2 mg. epinephrine in 1 cc. oil, corresponding to 0.6 to 0.7 mg. per kg. cat, does not produce a detectable increase of blood pressure. Only by massaging the site of the intramuscular injection is an increase of 7 to 32 mm. in the carotid blood pressure obtained when doses of 0.5 to 1 mg., or higher, are used.

Discussion. Injected in oily suspension, the effect of epinephrine on the blood pressure is noticeably diminished, while its delaying action on the absorption of penicillin remains extremely high. The latter effect, which depends on a local ischemic reaction, appears with doses of 0.05 to 0.1 mg., *i. e.*, about one-twentieth ot one-fortieth of the dose required for eliciting a systemic vasopressor action.

The duration of the blood level cannot be prolonged beyond a certain ceiling period (26 hours) by increasing the dose of epinephrine and penicillin above a definite amount. The "ceiling duration dose" for epinephrine is approximately 0.2 mg., while for penicillin it varies with the species. The existence of a ceiling dose for epinephrine is apparently related to the production of a *maximal* ischemic action on the surrounding tissue. It is more difficult to advance reasons for the "ceiling duration dose" of penicillin. This is probably determined by various factors: an end-point in the epinephrine effect, the dispersion of the oily material, the instability of penicillin. An additional factor might be represented by the rate of absorption of penicillin from the *deposit*. There is some justification for the assumption that the fraction of penicillin absorbed in equal time intervals does not change with the absolute amount of penicillin in the tissues, when given in oil epinephrine. Supposing that half of the penicillin present in the tissue is absorbed in 2 to 3 hours, the redoubling of the dose would cause an increase of the duration by 2 to 3 hours only.

With the larger *relative* doses required to produce an equal duration in the smaller species, the amounts of penicillin absorbed

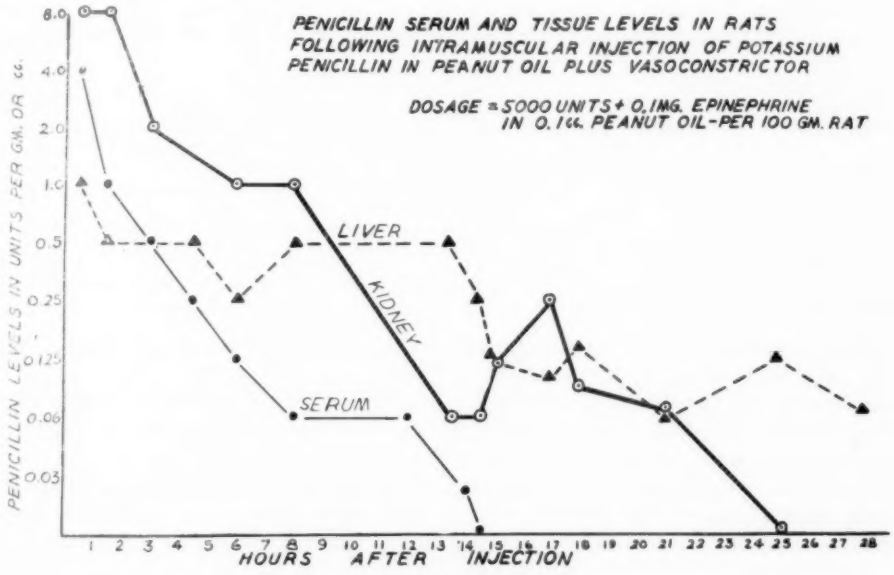
PENICILLIN SERUM AND TISSUE LEVELS IN RATS
FOLLOWING INTRAMUSCULAR INJECTION OF POTASSIUM
PENICILLIN IN PEANUT OIL PLUS VASOCONSTRICTOR
DOSAGE = 5000 UNITS + 0.1MG. EPINEPHRINE
IN 0.1cc. PEANUT OIL-PER 100 GM. RAT
PENICILLIN LEVELS IN UNITS PER GM. OR CC.
8.0
4.0
2.0
1.0
0.5
0.25
0.125
0.06
0.03
LIVER
KIDNEY
SERUM
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28
HOURS AFTER INJECTION

FIG. 4

during the same time period are correspondingly larger and this obviously brings about higher blood concentrations. In fact, the relation in the peak concentrations reached with a 300,000 u. dose, which gives a 1 day duration in man and in dog, is proportional to their weight differences: 0.5 to 1 u. in man, 5 to 20 u. in dogs. Similarly, an increase of dosage from 300,000 to 600,000 u. in man results in correspondingly higher blood levels. These differences in duration, according to species and relative weight, are important from a methodologic standpoint: one has to consider them when the prolongation obtained with a new preparation is interpreted.

of the initial high concentrations.

The therapeutic value of this "plateau" of high blood levels appears from the following consideration based on the clinical results of Cohn and co-workers in the treatment of gonorrhea. The active intramuscular dose which cures practically all (97%) cases of male gonorrhea is 0.5 cc. of our standard preparation corresponding to 150,000 u. of penicillin.[3] The "duration" of detectable blood level following such an injection is from 8 to 12 hours. Twice this dose, *i. e.*, 300,000 u. of penicillin injected in saline results, on the other hand, in an unsatisfactory cure rate,[4] though the duration of the *detectable* blood level is similar, 8 to 9 hours, and

TABLE 1.—PEAK SERUM CONCENTRATIONS IN DOG AND MAN FOLLOWING INTRAMUSCULAR ADMINISTRATION OF PENICILLIN IN SALINE AND IN PEANUT OIL PLUS VASOCONSTRICTOR

	Man			Dog		
Total dosage (units)	Units per kg.	Vehicle	Serum levels (units per cc.)*	Units per kg.	Vehicle	Serum levels (units per cc.)
200,000	2800	Saline	†	20,000	Saline	5.0–20.0
		Oil + Vasocon.	0.3–1.25		Oil + Vasocon.	1.2–5.0
300,000	4200	Saline	>1.25†	30,000	Saline	10.0
		Oil + Vasocon.	0.6		Oil + Vasocon	1.2–10.0
400,000	5700	Saline	†	40,000	Saline	10.0–20.0
		Oil + Vasocon.	0.6–1.25		Oil + Vasocon.	2.5–10.0

* The serum levels are the averages of a large number of determinations.

† In man the initial maximum concentrations in saline were not determined. The initial *transitory* peaks correspond to 4 to 5 units per cc.[9]

The experimental work on rats reveals that the prolongation of the penicillin in the organs, liver and kidney, outlasts by long periods the duration of the demonstrable blood level following injections in oil with vasoconstrictor. One might assume that the penicillin present in the organs—after its presence in the blood is no longer *detectable*—is of some therapeutic significance. A certain number of experiments carried out in this laboratory would indicate that the accumulation of penicillin in the organs, such as observed following treatment in oil with vasoconstrictor, cannot be obtained with high doses of penicillin given repeatedly in saline.

The experimental data suggest that the therapeutic advantages of the new penicillin preparation investigated depend on the protracted duration of the antibiotic in the tissues, and on the long "plateau"

the initial (very transitory) peaks are even higher.

Therefore, a comparison of the "duration" of blood levels with these 2 treatments does not provide a satisfactory explanation of the therapeutic differences. The curative effect of the 150,000 u. preparation, in contrast to the 300,000 u. given in saline, is due probably to the plateau effect at relatively high blood levels. This conclusion is in agreement with the data of Buck, Ercoli, Kelly, Lewis and Schnitzer[2] in experimental pneumonia: "the very high initial peak which was only observed after parenteral administration was of lesser significance than the consistent level of comparatively low concentration to be found in the serum during the hours following the subcutaneous as well as the oral administration."

The more prolonged retention of the

penicillin in the organs, as indicated by the urine levels, is probably an additional factor responsible for the increased therapeutic effectiveness of the suspension in oil with vasoconstrictor.

Summary. 1. The duration of the detectable penicillin blood level is not significantly increased when 200,000 to 300,000 u. are injected in oily suspension or emulsion, as compared to saline. The addition of very small doses of epinephrine to the oily suspension gives a noticeable increase in the blood level duration.

2. The systemic vasopressor activity of epinephrine is diminished by the oil, with the result that small fractions, one-twentieth to one-fortieth, of the therapeutic epinephrine dose prolong the penicillin levels.

3. Compared to the approximately *one day* duration of the blood levels, the urines of the subjects treated with 300,000 units of penicillin (in oil + vasoconstrictor) contained penicillin up to $2\frac{1}{2}$ to 3 days. In experimental animals, it was found that the treatment with this penicillin preparation gives about 2 times longer durations in the organs than in the blood.

4. The quantitative aspect of the blood level curves in relation to the dosage was studied. It is assumed that the improved therapeutic activity depends on the prolongation of the penicillin content in the organs and on the initial "plateau" levels in the blood.

5. Prophylactic experiments in mice infected with pneumococcus Type II indicate also an increased duration of therapeutic effectiveness of the penicillin preparation in oil and vasoconstrictor.

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SINGLE INJECTION TREATMENT OF GONORRHEA WITH POTASSIUM PENICILLIN SUSPENSION IN OIL-CONTAINING EPINEPHRINE

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Ever since the effectiveness of penicillin therapy of gonorrhea, by repeated administration of aqueous solutions of the drug was established in 1943,[6,7] numerous attempts have been made to reduce the treatment to a single injection.[9] Various vehicles known to prolong the duration of penicillin effect have been used for this purpose.[4,10] The water-in-oil emulsion proposed by Freund and Thomson[5] gave, in our experience[1] satisfactory therapeutic results as did the beeswax-in-oil mixture according to Romansky.[11]

These preparations have, however, a certain number of shortcomings for the practitioner. The beeswax mixture is thick and becomes a resinous uninjectable mass if any moisture reaches it. The beeswax occasionally produces foreign body reactions and local abscesses.[11] The practical disadvantage of the material previously studied by us, *i. e.*, the penicillin water-in-oil emulsion, is that the emulsification process has to be done by the physician himself, with sterile materials, and the emulsion requires refrigeration.

The mixture used in the present clinical investigation proved satisfactory in overcoming the disadvantages of the preceding preparations. It consists in a suspension of crystalline penicillin-in-oil containing epinephrine. It is stable at room temperature and after brief shaking of the ampoule is ready for aspiration and injection: it flows readily. The data relating to the prolongation of blood levels in animals and man were presented by Ercoli, Hueper, Landis, Queally and Schwartz.[3] We treated a total of 300 cases of male gonorrhea with this preparation. The therapeutic results are reported in this communication.*

I. Diagnosis, Treatment and Determination of Cure. The cases treated were 300 consecutive male ambulatory patients with acute gonococcic infections. The diagnosis was established by routine bacteriologic examinations of urethral smears and cultures. The preparation was injected intramuscularly into the buttocks or subcutaneously into the upper outer quadrant of the gluteal region. For the major portion of the investigation an ampoule containing 300,000 units of potassium penicillin and 0.3 mg. of epinephrine in 1 cc. of a vegetable oil was used. The doses injected varied between 0.25, 0.5 and 1 cc. Only exceptionally were 2 cc. doses used.

The criteria of cure were the following: (1) the absence of urethral discharge; (2) clear urine in the "2 glass" tests; (3) at least 2 negative cultures of both urethral and prostatic secretions during a period of at least 2 weeks (37 cases). In the majority of the cases followed up completely (117 out of 154), 3 prostatic cultures were taken during a period of 3 weeks. Peizer's medium was used for cultures; the identification of gonococcus was confirmed in each case by sugar fermentation tests.

II. Therapeutic Results. Out of 300 patients injected with various doses

* The material used in this study was made available to us by the courtesy of Warner Institute for Therapeutic Research, New York, N. Y.

of the preparation investigated, a total of 154 could be followed up for 2 to 3 weeks, for the final determination of cure. The remaining 146 cases were delinquent (48.7%). The results of treatment with different dosage are given in Table 1.

TABLE 1.—SINGLE INJECTION TREATMENT OF GONORRHEA WITH A SUSPENSION OF PENICILLIN IN OIL CONTAINING EPINEPHRINE

Volume injected (cc.)	Epinephrine (mg.)	Penicillin (Oxford units)	No. cases	No. cured	No. failed
		Intramuscular Administration			
0.25	0.06-0.15	75,000	2	1	1
0.50	0.15-0.50	150,000	45	44	1
1.00	0.1	200,000	19	19	0
1.00	0.3	300,000	7	7	0
2.00	0.6	600,000	4	4	0
		Subcutaneous Administration			
0.25	0.06-0.15	75,000	7	6	1
0.50	0.15-0.50	150,000	54	52	2
1.00	0.25-0.30	300,000	11	11	0
2.00	0.60-2.00	600,000	5	5	0
Grand total	...	...	154	149	5

It appears that a single injection of 0.5 cc. of the oily suspension, containing 150,000 units of penicillin and 0.15 mg. of epinephrine is sufficient to cure gonorrhea infection in the male. The bacteriologic and clinical examination of 100 patients so treated and followed up for 2 to 3 weeks indicated a cure rate of 97%. Two of the 3 cases which relapsed showed transitory negative bacteriologic findings for a period of 24 hours and relapsed soon thereafter. Since these patients definitely denied any exposure, a diagnosis of relapse was made.

A single injection with 0.25 cc. of the suspension containing 75,000 units and 0.075 to 0.15 mg. of epinephrine cured 7 out of 9 patients. In spite of the small number of patients in this group, it is justifiable to conclude that this dose is insufficient for therapy. A single injection with 1 or 2 cc. of the suspension containing from 200,000 to 600,000 units cured all 46 cases followed up.

It is noteworthy that 3 cases which relapsed following treatment with 0.5 cc. per 150,000 units dose responded promptly to a second injection of the same dose. One case of relapse following the administration of 0.25 cc. per 75,000 units dose was successfully treated with 0.5 cc. per 150,000 units, given by single injection. (The last case of relapse following the 75,000 unit treatment was delinquent.)

After probable cure and admitted sex exposure, 9 cases were diagnosed as reinfections. Five of these cases were retreated with the original dose used, *i. e.*, 0.5 cc. per 150,000 units in 4 cases and 1 cc. per 200,000 units in the fifth case. All these patients were promptly cured. The 4 remaining cases were delinquent.

Immediate reaction to intramuscular injections of the suspension were similar to those observed when aqueous penicillin solutions are injected. They consist chiefly of a transient burning painful sensation which began about 20 seconds after the injection and lasted for about 2 to 3 minutes. Sometimes it radiated downwards, to subside gradually within 5 minutes. Subcutaneous injection produced no radiating pain, but in some cases a transitory 2 to 3 minutes' painful burning sensation was felt. The site of injection was examined repeatedly during the course of the follow-up period and no local infiltration or other abnormalities were found except in 2 patients who developed 3 cm. subcutaneous indurations which lasted for about 28 days. One of these patients was treated with a very high dose of epinephrine; 2 mg. in 2 cc.

Among 15 patients whose blood pres-

sures were taken before and after the administration of varying amounts of the mixture containing epinephrine, 8 showed no rise or fall in blood pressure. Six patients showed a drop up to 16 mm. of mercury within 15 minutes of the injection and 1 a rise of the blood pressure up to 14 mm. of mercury. Thus, there was no significant effect on the blood pressure. There was no rise in the pulse rate. There were 2 cases which showed exacerbations of a preëxisting dermatophytosis and *Tinea cruris*.

Discussion. The foregoing observations indicate that the minimal effective dose for the treatment of gonorrhea by single injection with a preparation of penicillin in an oily suspension containing vasoconstrictor, consists of 0.5 cc. of the oily suspension containing 150,000 units of crystalline penicillin and 0.15 mg. of epinephrine. A cure rate of 97% followed this dosage. A second injection with the same dosage likewise cured the 3 relapsing cases. There were no relapses in the 46 cases treated with doses higher than 150,000 units.

Our present results of therapy compare favorably with those following the administration of 150,000 units of penicillin in 4.5 cc. of a water-in-oil emulsion (cure rate 96.2%)[1] and with multiple injections of penicillin in aqueous solution (cure rate, 96.5 to 100%).[2]

The prolongation of the penicillin effect by using epinephrine-in-oil as a vehicle for penicillin[3] is probably responsible for the excellent therapeutic results. Comparing our clinical findings with the figures on blood levels given by Ercoli, Hueper, Landis, Queally and Schwartz, it would seem that the therapeutic dose of 150,000 units yields detectable amounts of penicillin in the serum of the patient for a period of approximately 12 hours. The observation that the relapses are as sensitive to treatment as the initial infection cannot be explained in the light of our present knowledge. It indicated, in any event, that the relapse strain is certainly not becoming resistant to penicillin. This observation limited to a small, statistically insignificant number of cases deserves further study for its broader significance of the relation of penicillin effect upon a relapsing infection.

Summary and Conclusions. 1. This study was directed toward the evaluation of a single injection treatment of gonorrhea with a suspension of potassium penicillin in oil containing epinephrine.

2. Single injections of 150,000 units intramuscularly or subcutaneously, in a total volume of 0.5 cc. cured 97 of 100 patients. The 3 patients who did not respond to the initial injection were promptly cured with a second injection of an identical amount of penicillin.

3. All of 19 patients who were treated with 200,000 units in 1 cc. intramuscularly were cured. Likewise all of 18 patients treated with 300,000 units in 1 cc. and all of 9 patients treated with 600,000 units in a total volume of 2 cc. responded to this form of therapy.

4. No untoward local or systemic reactions were observed following the subcutaneous or intramuscular injection of the suspension of crystalline potassium penicillin in oil with epinephrine.

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THE EFFECTIVENESS OF PENICILLIN IN THE TREATMENT OF NASOPHARYNGEAL DIPHTHERIA

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The early investigation of penicillin for clinical use revealed its effectiveness against the *Corynebacterium diphtheriæ*. The efficacy of penicillin in the treatment of diphtheria has not been as fully studied as in other diseases. Furthermore, controversy still exists concerning the clinical value of penicillin in diphtheria. The comparative paucity of clinical material has been partly responsible; the presence of the diphtheria toxin has also complicated the problem.

This paper deals only with the management of the early case of nasopharyngeal diphtheria. From the data presented, it is thought that some conclusions can be drawn regarding the efficacy of penicillin treatment. It must be realized that all the patients studied had been previously immunized according to the prescribed Army Schedule.[10] Initial immunization had been completed on all cases included, but the dates thereof and the dates of "booster" injections are not known.

Procedure. The case material was collected over an 8 month period, from September 1946 to May 1947. The patients in this series were young men in the United States Army occupation forces deployed in the general area surrounding Frankfurt a/M, Germany. The hospital in which the patients were studied was the 97th General Hospital located in Frankfurt.

Diagnosis. All cases diagnosed as diphtheria (nasopharyngeal) had the usual history of acute sore throat, though varying greatly in intensity. General malaise and systemic complaints were usually present. The physical examinations revealed evidence of nasopharyngitis. Briefly, there was usually erythema and congestion of nasal passages. The pharynx, palate, tonsils, if present, and surrounding tissues showed erythema and edema. The close-fitting, smooth, gray membrane described as typical of diphtheria was seldom seen. Every case herein reported, however, had a positive nasal and/or pharyngeal culture for *Corynebacterium diphtheriæ;* and further, each culture reported positive and included in this series had a confirmed positive virulence test. The severity of the physical findings could not be correlated with positive diphtheria culture reports, since many patients had a coincident follicular or parenchymatous tonsillitis.

Bacteriology. Bacteriologic identification of the corynebacterium was done routinely by the laboratory. The standard media and identification procedures in use throughout Army laboratories were employed here.

Every culture positively identified was forwarded to the United States Army 4th Medical Laboratory. Here virulence studies were done under standard technique employing guinea pigs. As mentioned above, only those cases giving positive virulence studies are included.

Summary Card. To simplify the following of patients and assembling data, summary cards were used. These had been in use by the medical service at the hospital for some time previous to this study. Since they proved to be of much value, a sample is shown in Figure 1.

A total of 139 cases are presented; 88 of these received the "routine" treatment; 51 received the "routine" treatment plus "penicillin early."

Type of Treatment. (*a*) By "routine treatment" is meant the following: Bed rest, nursing care, necessary sedation and analgesia, special diets and the usual general medical measures. The specific or local treatment consisted of mouth washes and gargles prepared of warm water, saline and occasionally peroxide. Every case received diphtheria antitoxin 100,000 units intramuscularly. The antitoxin was given in each instance as soon as a positive culture for

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corynebacterium was reported. In some instances the antitoxin was given before the culture report was received. The report of the virulence test was not awaited before giving the antitoxin because of the additional time interval involved. In the great majority of the patients sulfadiazine had been used before a diagnosis of diphtheria was established. Exact figures on the number of patients receiving sulfadiazine are not known, since the special reference cards did not always include that information. However, the distribution of patients treated with sulfadiazine is approximately equal in both groups. No penicillin was given as a routine measure, either locally or systemically.

2 days, and at 4 weeks plus 4 days. If the last 3 cultures taken were negative, the patient was considered no longer a carrier and was transferred for convalescence or discharge. If any of the cultures following the original one were positive, the patient received further treatment as outlined below. Following this second positive report, again a period of 2 weeks elapsed before the series of 3 cultures 2 days apart was repeated for clearance.

Results. (*a*) *Routine Treatment.* Of the 88 cases treated according to the "routine" method, 46 showed a positive culture at the first 2 week interval; 35 cases showed a negative culture at the

FIG 1.—SAMPLE PATIENTS' SUMMARY CARD OF CLINICAL DATA

NAME	Date of 1st positive culture	Dates for future cultures	Electrocardiogram	Treatment	Remarks	Type of organism
.....	14 Jan. N/P —neg./pos.		14 Jan. —normal	Penicillin, 20,000 u. q. 3 hrs. 14 Jan.–20 Jan.		Gravis
			21 Jan. —normal			
.....		28 Jan. N/P —neg.	28 Jan. —normal		Up and about	
.....		11 Feb. N/P —neg.				
.....		13 Feb. N/P —neg.			No complaints	
.....		15 Feb. N/P —neg./pos.	15 Feb. —T wave change	Tonsillectomy, 17 Feb.		Gravis
.....			22 Feb. —normal	Penicillin, 20,000 u. q. 3 hrs. 16–18 Feb.		
.....		1 Mar. N/P—neg.	1 Mar. —normal		Up and about	
.....		3 Mar. N/P—neg.			No complaints	
.....		5 Mar. N/P—neg.			Discharged	
.....	.....	.....	.....	.....	.....	.....
.....	.....	.....	.....	.....	.....	.....

N—nasal culture. P—pharyngeal culture.

(*b*) By routine plus "penicillin early" is meant the following: All the measures mentioned above were included; but in addition the patient received penicillin intramuscularly. This drug was not used locally. And it should be noted that the penicillin was begun at the first report (bacteriologically) of a positive culture for corynebacterium. These reports were usually available within 4 days. The penicillin was given intramuscularly, 20,000 units every 3 hours. This was continued for 50 doses or 1 million units, unless otherwise stated. This dosage was arrived at empirically and with the advice of others studying the problem.

Treatment Schedule. Following a positive culture report for *C. diphtheriæ*, the patient was transferred to the isolation section. Cultures of the nose and throat were then taken at an interval of 2 weeks from the original positive culture, at 4 weeks, at 4 weeks plus

2 week interval; 7 additional cases treated in the routine manner, negative at the 2 week period, later were shown to be in a carrier state by a positive culture appearing in the clearance series. Thus a total of 53 cases (60%) developed a carrier state as opposed to the 35 (40%) who did not.

(*b*) *Penicillin Early.* Of the 51 cases who received the routine plus "early" penicillin treatment, 43 had negative cultures at the 2 week interval and 8 were positive. Of the 43, only 3 who were negative at 2 weeks, later became positive and required further treatment. The (79%), with permanently negative cultures as opposed to 11 positive cases (21%) is obviously a better result than that of those getting "routine treatment."

It may be mentioned here that the 2 week culture seems to be of considerable value as an aid to recognizing the possible development of the chronic carrier state. This is supported by the fact that only 10 (13%) of the 75 cases negative at 2 weeks, later became positive. In other words, prognosis regarding the absence of the carrier state is nearly 90% correct when based on the 2 week culture alone. This figure becomes even higher when the "early" penicillin group is considered alone. As mentioned, only 3 out of 43 cases first negative later became positive. This is less than 7% error.

Further evaluation of the above is not possible because the patients reported positive at the 2 week interval were put on intensified treatment programs immediately. This was done to shorten the period of hospitalization necessary.

(*c*) *Hospitalization Time.* A comparative study of the required time for hospitalization between the 2 series is of interest (Table 1). The average time of hospitalization for patients with a negative 2 week culture was 35 days; and the average time of hospitalization with a positive 2 week culture was 50.3 days. These figures for average hospitalization include patients who became negative without further treatment after a 2 week positive culture. They do not include some of those patients held beyond 50 days for recovery from complicating carditis. These patients had positive cultures at the 2 week interval. Consequently the 50 day figure is in error on the low side only. This difference in average hospitalization is also significant. These data give additional support to the value of the 2 week culture.

(*d*) *Penicillin Late.* In contrast to the results with penicillin given early, the late use of penicillin intramuscularly was not so successful; 40 cases of the positive 2 week group (53) treated routinely on admission, received penicillin only when the 2 week culture had been reported positive. They received 1 million units of penicillin intramuscularly, 50 doses of 20,000 units at 3 hour intervals. Of these 40 cases, 16 showed negative cultures (40%), while 24 (60%) remained positive. Several of these 24 cases received penicillin in the dosage above a second time. None was successfully cleared of the carrier state until tonsillectomies were performed.

COMPLICATIONS. There were no serious complications seen in this series. The most frequent postdiphtheritic damage found was a carditis diagnosed only by the electrocardiogram. Of the "routine" and penicillin "early" cases responding with a negative culture series, the percentage showing carditis beyond the first 2 weeks was approximately the same; namely, 42% for the former and 40% for the latter.

Of the unsuccessful group treated routinely, that is, those positive at 2 weeks, 19 of 54 cases (35.2%) showed a carditis. Certainly from this point of view penicillin cannot be said to have helped. However, of the group who underwent tonsillectomies, following penicillin at 2 weeks to clear the carrier state, 50% showed continuing electrocardiographic changes for more than 4 weeks. The other 2 groups mentioned above (those with negative 2 week cultures) each showed only 30% having electrocardiographic changes longer than 4 weeks.

The same group undergoing tonsillectomies had the longest average hospitalization of over 61 days. This suggests that the longer an organism remains in the throat, the greater its chance of doing damage as manifested in this series by continuing changes in the electrocardiogram.

Discussion. It was interesting to find that the majority of cases harbored the gravis strain. This was almost universally true until November 1946. From that time on, a gradually increasing proportion showed the mitis strain. By March 1947 over 50% of cases were of mitis etiology. There was no significant difference found in the incidence of complications when cases due to the mitis strain were compared to cases due to the gravis strain.

Shick tests were not done on these patients.

It should be mentioned that every case undergoing tonsillectomy received penicillin (20,000 units every 3 hours) for 24 hours preceding operation until 24 hours after the operation. In persistent carrier states this therapy was most effective in clearing the throat of the organism. The data on results of treatment are summarized in Table 2.

severe cases. The absence of severe complications is also attributed to the early administration of diphtheria antitoxin. Only 3 cases seen during this entire period showed severe complications. These 3 are not included here because they were primarily skin diphtheria cases.* None of the 3 received antitoxin for from 3 to 4 weeks after the onset of skin lesions because the diagnosis was unsuspected. In each case a severe peripheral neuritis

TABLE 1.—COMPARISON OF TREATMENT AND HOSPITALIZATION

Cases	Treatment	Negative at 2 wks.	Positive at 2 wks.
88	Routine	35 (40%)	53* (60%)
51	Penicillin "early"	40 (79%)	11 (31%)
38*	Penicillin "late"	16 (40%)	24 (60%)
	Average days hospitalization	35	50 3

* Continuation of same cases.

TABLE 2.—FINAL DISPOSITION OF ALL CASES

Cases			Negative at 2 wks.	Positive at 2 wks.			Negative at 2 wks.	Positive at 2 wks.			Negative at 2 wks.	Positive at 2 wks.
139		Treatment				Treatment				Treatment		
51	51	Penicillin "early"	40	11	6	Tonsillectomy	6					
					4	Time*	4					
					1	Spray-penicillin	1					
88	88	Routine	35	53	40	Penicillin "late"	16	24	22	Tonsillectomy	22	
									2	Time*	2	
					6	Tonsillectomy	5	1	1	Time*	1	
					2	Tonsillectomy plus 1 million penicillin u.	2					
					2	Time*	2					
					3	Spray-penicillin	3					
139			75				59				25	
Average days hospitalization			35	50							61	
% case groups showing ECG change over 4 wks.			30	30							50	

* See discussion.

As can be seen, only 4 cases received penicillin by nasopharyngeal spray. The results here confirm those of Kocher and Siemson,[3] who found nose drops and lozenges of penicillin effective in 74% of 31 carriers within 7 days. These results supported earlier work by Turner.[9]

Any general clinical response due to penicillin could not be evaluated. Systemic symptoms and signs cleared rapidly in all cases.

It is believed that previous immunization was responsible for the absence of

set in. Complete follow-up is not available. From observation, it is known that approximately 2 to 3 months elapsed before improvement was noticeable.

The above results give support to the conclusion (Karelitz[2]) that penicillin given early is effective in preventing the development of the carrier state. In so doing it decreases the hospitalization time for diphtheria patients. No statements can be made regarding the efficacy of higher doses of penicillin as recommended by Long[6] and Skinner.[8] The data assembled

* Study to be published in Arch. Derm. and Syph.

from the mild complications observed are in agreement with Weinstein,[11] who doubts that they can be prevented by penicillin alone. In this regard early administration of diphtheria antitoxin is imperative.

The pathogenesis of cutaneous diphtheria is closely associated with a lowered skin resistance.[4,5] Existing cutaneous infections is a frequent precursor of invasion by cutaneous diphtheria. Similarly in nasopharyngeal diphtheria the original damage may be done by the so-called secondary invaders invariably found present. The stage is thus set for invasion by the corynebacterium. Since penicillin is effective against the most common pharyngeal secondary invaders, it will be effective in the type of case seen here and rapid healing of the pharyngeal membrane will be promoted. The local tissue reaction is then able to destroy the corynebacteria before seeding is completed to develop the carrier state. Long[6] has observed that when clinical improvement due to penicillin is pronounced, secondary invaders are prominent. Action of penicillin against *C. diphtheriæ* is probably also effective, particularly in increased dosage.[6,8,11] This is known to occur *in vitro*.

Patients developing the carrier state usually have tonsils present and/or considerable pharyngeal lymphoid tissue. Here the local use of penicillin can be expected to be of more value than the intramuscular method. Tonsillectomies should be done where indicated.

A study of nose and throat cultures for secondary invaders during infection with cutaneous diphtheria might be revealing. Response to the appropriate chemotherapy as indicated by the cultures could include penicillin, sulfa drugs or other specifics.

Summary. 1. In a series of 139 cases of diphtheria, treatment with penicillin was followed by a decrease in the incidence of carriers, if given early in the course of the disease.

2. There is also a shorter period of hospitalization for early penicillin treated cases than for others.

3. The incidence of postdiphtheritic complications was not affected by penicillin; however, the severity of complications was modified.

4. A suggestion is made as to the possible mode of action of penicillin.

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A CLINICAL EVALUATION IN CHILDREN OF THE TOXICITY AND EFFICACY OF CARONAMIDE (4-CARBOXYPHENYLMETHANESULFONANILIDE) FOR THE COMPETITIVE INHIBITION OF PENICILLIN EXCRETION*

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In recent publications by Beyer and his co-workers[1] it was demonstrated that caronamide (4-carboxyphenylmethanesulfonanilide) administered orally, by inhibiting the rapid renal excretion of penicillin, was capable of maintaining higher and more sustained levels of penicillin than normally occurred after a given dose. In this research there is presented an evaluation of the toxicity of caronamide in children and of its efficacy in increasing the plasma concentration of orally administered penicillin.

Penicillin is lost rapidly from the body fluids because of its rapid renal elimination. It has been shown that penicillin is excreted by the kidney in the same manner as is p-aminohippuric acid (PAH). Since PAH at low blood concentrations is almost completely extracted from renal arterial blood in one circulation through the kidneys, it follows that the amount of PAH appearing in the urine per minute is practically equal to the quantity of PAH contained in the volume of plasma circulating through the kidney per minute. Thus the renal clearance for PAH and penicillin (*i. e.* the quantity of plasma from which these substances are completely removed in one minute by the kidney) is a measure of the renal plasma flow per minute.[2] The renal clearance of penicillin is accomplished by a combination of glomerular filtration and tubular excretion. Since this clearance approximates the renal plasma flow and since in man 20% of the renal plasma flow is filtered at the glomerulus (the so-called filtration fraction) the greater part (80%) of urinary penicillin is excreted by the tubules.

Caronamide decreases the rapid renal excretion of penicillin by its inhibitory action on the tubular excretory mechanism for penicillin. This inhibition is effected by halting the tubular transport mechanism involved, without caronamide itself being excreted by the tubules and without inhibiting other tubular transport mechanisms.[3] This compound is eliminated essentially by glomerular filtration. The inhibitory effect of caronamide on the tubular transport mechanism is analogous to the *in vitro* competitive inhibition of the action of an enzyme on its substrate by another compound possessing an affinity for, but being refractory to, the action of the enzyme.

The mode of action of caronamide may be contrasted with the inhibition of penicillin excretion by Diodrast or PAH

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(p-aminohippurate), which are excreted by the same mechanism as is penicillin.[8] These latter agents do not suppress the function of that common transport system but in being excreted by it limit its functional capacity to excrete penicillin on a "mass action" basis. The rapid tubular excretion of PAH and Diodrast, together with the necessity for the parenteral administration of either agent, make their use for this purpose impractical in ordinary penicillin therapy. Caronamide, because it is excreted only by glomerular filtration, is eliminated at a relatively slower rate and may be administered orally.

The compound caronamide was described by Sprague, Ziegler, Miller and Cragoe.[10] It has the following chemical structure:

Beyer[1] has reported this agent (1) to fulfill the original physiologic limitations imposed by the concept of its mode of action; (2) to be markedly effective in dogs in inhibiting the renal tubular elimination of penicillin at moderate dosages and in a reversible manner; (3) to enhance from 4- to 16-fold the chemotherapeutic effect of penicillin in experimental pneumococcic and typhoid infections in mice;* and (4) to have low order or toxicity in the mouse, rat, rabbit, dog and monkey.

Toxicity Studies. Three groups of 6 patients each were selected from the orthopedic and medical services of the Children's Hospital of Philadelphia for this phase of the work. These children constituted a heterogeneous group from the standpoint of diagnosis and present medical histories. Fifteen patients were studied; three of them were used in both the first and third group. Ages ranged from 2 to 13 years and weight from 21 to 100 pounds (9.5 to 45.5 kg.).

Transposing from the experimentation on dogs, it was anticipated that an effective dosage of the compound would be about 0.2 gm./kg./24 hours. Consequently the 3 groups of patients were given total caronamide dosages of 0.1, 0.2 and 0.4 gm./kg./day, divided into equal aliquots and administered at intervals of 4 hours, day and night. The duration of each group study was at least 7 days. Before beginning each study, 24-hour urine collections and fasting blood samples for chemical and hematologic examination were obtained from each patient. Chemical and hematologic analyses were performed again after the first 3 or 4 days of drug administration and finally at the termination of the experiment.

A systematic daily record, except for hematology and chemistry, wherein the following data were recorded, was kept for each patient: subjective and objective manifestations of toxicity such as nausea, vomiting, rash; daily temperature range; appetite; weight; 24-hour fluid intake and output; and blood pressure. In addition the following data were obtained: total erythrocyte and leukocyte counts; differential white cell counts; hemoglobin; blood urea, glucose, N.P.N.; urine casts, crystals, microscopic hematuria, proteinuria, reducing substances, total nitrogen, Gmelin's test for bile pigments, specific gravity, and pH.

The course of events was such that the group receiving the 0.1 gm./kg./day dosage was started first. No patient was given less than 3 gm. per day, 0.5 gm. every 4 hours, even if the calculated dose was less than this amount. After 3 days wherein no sign of drug tolerance was evident, the second group was started on a dosage of 0.2 gm./kg./day. After 4 days, the third group was begun, and this included 3 children from the first group whose medication was interrupted for only 2 days. The daily dosage for this group was 0.4 gm./kg./day. Administration of the drug to 3 children in this group was stopped at the end of one week, but the others were allowed to continue taking it

* Detailed evidence in support of this statement is given in an article by Verwey and Miller (Proc. Soc. Exp. Biol. and Med., **65**, 222, 1947).

for a second week. Since the drug was available in 0.5 gm. tablets, in the case of the smaller children, they usually were mashed in a spoon and administered with orange, cherry, or strawberry syrup.

RESULTS. There were no manifestations of toxicity that necessitated withdrawal of caronamide from any of these 15 patients or the next group of 5 that received penicillin plus the drug. The patients tolerated the caronamide well. There was only one instance of vomiting of the tablets from day to day, and that was in a patient who had tolerated 6 gm. per day but who vomited frequently when given 18 gm. per day as a member of the third group. Other isolated instances of vomiting were attributable to giving the tablets without water, to a meal that the child disliked, or to other similar secondary causes.

There were 2 isolated instances of crystalluria among the repeated urinalyses on each patient. These occurred in the second group (0.2 gm./kg./day) and at a urinary pH of 5.9 and 6.1 respectively in the 2 patients. These 2 children received 5 and 5.5 gm. of the drug per day. The crystals were clustered as sheaves. There seems reason to believe that the development of crystalluria was dependent on the pH of the urine, since it was not encountered at pH values above 6.2. However, there were 9 instances in which the urinary pH was below 6.2, in one case 5.9, where crystalluria was absent. No attempt was made in this study to avoid or attempt to correct these transient findings. The crystallurias were not accompanied by gross or microscopic hematuria or by casts.

There were no changes in hematologic findings or persistent elevation of temperature attributable to the drug. To be sure, temperature and leukocyte count increased following an intercurrent pharyngitis, the development of a wound infection, and following a smallpox vaccination. The criterion of a drug fever was taken as an otherwise unexplained elevation of rectal temperature to over 100° F. on at least 2 successive days any time during caronamide administration.

There were 3 instances of the appearance of a rash during or immediately following the administration of the drug. No 2 cases were alike in the nature or distribution of the rash. The first occurred on the fourth day of drug administration, the patient being in the first group (0.1 gm./kg./day). This rash consisted of small erythematous, grouped papules limited to the extensor surfaces of both wrists and was preceded by itching several days prior to the appearance of the rash. This child began to use crutches with leather arm straps that were in constant moving contact with the wrists and it was felt that the rash represented a contact dermatitis rather than a drug reaction. Only traces of the rash could be made out at the termination of the experiment 3 days later. On the fifth day a second patient in the third group (0.4 gm./kg./day) developed a moderate maculo-papulo-vesicular rash on the lower portions of the legs and feet, including the soles. There were a few lesions on the lower portions of her arms. This faded almost entirely within 24 hours. In both of these instances the lesions faded despite the continued administration of the drug.

The third case occurred in the patient in the third group previously mentioned as having vomited frequently. She developed a florid small macular rash over her face, neck, arms and chest 24 hours after the termination of drug administration, which was accompanied by a mild palpebral conjunctivitis and a mild leukocytosis. There was no adenopathy. The rash faded completely within 48 hours after its appearance. All 3 children who manifested these dissimilar rashes were convalescent cases of poliomyelitis.

Another interesting observation was the appearance of reducing substance in the urine of 3 patients in the third group, one of them being the patient described as having a rash 24 hours after discontinuation of the drug. In this case and in another the reducing substance was pres-

ent on only one occasion, but in a third patient it was present consistently during drug administration.

This reducing substance was not glucose, nor was there any change in the blood glucose level at the time of its appearance or later. Instead, it appeared to be a pentose. In each instance where a qualitative test for urinary reducing substance was positive (Benedict's Test) it was also estimated quantitatively. A similar amount of glucose was added to normal urine and aliquots of both specimens were yeasted to remove fermentable substances. Following yeasting, the control urines were negative whereas the test samples still contained their initial concentration of reducing substance. Tests for glucuronates were negative. Lactose was not the nonfermentable substance. The case records for medication, diet, etc., were irrelevant.

It is probable that these were instances of pentosuria. One case of pentosuria (if such it was) occurred only at the time of a pharyngitis in a convalescent poliomyelitis patient. Another occurred following vaccination against smallpox of a patient having congenital malformation of the joints. The pentosuria was associated with an elevation of temperature. In the third case the pentosuria was only detected on the last day of the drug administration, 24 hours before the appearance of a rash. It is quite possible that the pentosuria was precipitated by the drug under conditions of metabolic stress in patients undergoing some abnormal pattern of muscular or joint metabolism. A certain amount of credence may be lent this interpretation since pentosuria only occurred in post-poliomyelitic cases with the exception of 1 case, involving congenital malformations of the joints necessitating the patient's legs being in a cast.

This finding recalls the observation of Dr. A. S. Minot that patients having progressive muscular dystrophy have pentosuria.[7] In contrast to this explanation was the report by Beyer, McKinney, and Tillson that young dogs given 1 to 1.5 gm. of caronamide/kg./day on an identical 4-hour dosage régime developed pentosuria. Dogs given smaller daily dosages of the drug did not manifest pentosuria.[4c]

The occurrence of non-fermentable reducing substance in the urine has been the first and usually the only characteristic systemic effect of the drug. The pentosuria was reversible by withdrawal of the agent, and its continued occurrence seemed of little consequence in either our patients or in the dogs referred to above.

In general there were no effects attributable to drug reaction that have not been noted in the foregoing discussion.

The Effect of Caronamide on Renal Function and the Blood Level Response to Penicillin Administered Orally. The design of these experiments was as follows: Five patients were selected for the study. On successive days before administration of penicillin and caronamide renal function tests were performed on each patient. These included measurements of the glomerular filtration rate (manitol clearance), minimal renal plasma flow (PAH clearance at low plasma levels), maximal ability of the tubules to reabsorb glucose (glucose Tm) and the maximal capacity of the tubules to excrete p-amino hippurate (PAH Tm).

Following the renal function test the first penicillin control phase wherein each patient was given orally 100,000 units of crystalline sodium penicillin-G* dissolved in water immediately before administration and flavored with a fruit syrup was begun. The dosage was repeated every 4 hours day and night. Each morning the 9 A.M. dose was given by one of us (other dosages were administered by the nursing staff). Breakfast was withheld until a blood sample was drawn exactly 1 hour after the drug was given.

The duration of the first control phase was 4 days. The schedule was so arranged

* The sodium salt of crystalline penicillin-G used in this work is thought to be at least 90% pure. It was obtained from the Commercial Solvents Corp., Terre Haute, Ind.

that during this period one 4-hour penicillin dose—plasma concentration response curve was taken on each patient. These were always started after the 9 A.M. dosage and all food was withheld until after the last or 4-hour blood sample was taken for penicillin assay. Thereafter, at 1 P.M., the next penicillin dose and lunch were given the patients.

Since 10 cc. samples of citrated blood were desired for the Rammelkamp assays, the experiment had to be designed to minimize bleedings and yet obtain a maximal amount of information. Sufficient blood was drawn to permit repetition of an assay when desired. The bloods were chilled immediately and were handled aseptically throughout. The organism used in the assay of the plasma was a hemolytic streptococcus. Human red cells were used in the determination of end points and a half-step dilution system was employed to minimize the percentage error of the assay.[5] It had been ascertained previously that caronamide did not influence penicillin assays.[4a] The lower limit of the assays was 0.02 μ./cc.

Immediately following the control period the first drug phase, wherein a daily dosage of 0.2 gm./kg. of caronamide was divided as equally as possible into 6 aliquots, and administered every 4 hours together with 100,000 units of penicillin, was begun. Here again the duration of the test was 4 days, penicillin blood levels were determined daily 1 hour following the 9 A.M. dose, and a 4-hour penicillin blood level curve was obtained on each patient, once during the period.

Following the initial drug phase the daily dosage of caronamide was doubled (0.4 gm./kg.). Otherwise the protocol was as has been outlined for the first drug phase. After the second 4-day drug period the administration of caronamide was stopped.

The administration of penicillin was continued as a second control phase. Forty-eight hours after the last dose of caronamide and on 2 succeeding days either a 1-hour, or a 4-hour, penicillin blood level curve was obtained on each patient. There were 2 exceptions to the 1-hour bleedings following the 9 A.M. dosage where this would have conflicted with renal function procedures. The renal function tests were repeated on the children during and following this second penicillin control period. The tests on the first child were performed 48 hours after the last dose of caronamide.

Throughout this whole procedure the laboratory and clinical studies, except for chemistry, were carried out periodically as in the toxicity studies. There were no notable changes disclosed by these tests.

Results. The general effect of caronamide on the blood level response to penicillin administered orally has been summarized in Table 1. Herein all the control values and blood levels at both levels of caronamide dosage have been averaged and compared at corresponding time intervals following the 9 A.M. dose of 100,000

TABLE 1.—THE AVERAGE PENICILLIN PLASMA LEVEL RESPONSE OF 5 PATIENTS TO THE ORAL ADMINISTRATION OF THE ANTIBIOTIC AGENT ALONE AND WITH CARONAMIDE

The dosage of penicillin was 100,000 units every 4 hours. The dose of caronamide administered with each dose of penicillin was one-sixth the daily dosage of 0.2 gm./kg./day or of 0.4 gm./kg./day.

Time after dose	Penicillin control (plasma conc. u./cc.)	Penicillin plus caronamide (0.2 gm./kg./day)		Penicillin plus caronamide (0.4 gm./kg./day)	
		Plasma conc. (u./cc.)	Fold increase over control	Plasma conc. (u./cc.)	Fold increase over control
1 hr.	0.325 (28)*	0.507 (20)	1.6	0.916 (20)	2.8
2 hr.	0.082 (10)	0.155 (5)	1.9	0.412 (5)	5.0
3 hr.	0.032 (10)	0.090 (5)	2.8	0.257 (5)	8.0
4 hr.	0.013 (10)	0.026 (5)	2.0	0.188 (5)	14.5

* Number of samples.

units of penicillin, or of penicillin and drug.

Scanning the data in Table 1, it is apparent that the overall effect of the 0.2 gm./kg./day dosage was to increase the penicillin plasma concentration 1.8- to 2.8-fold over the control values. The 0.4 gm./kg./day dosage of caronamide further increased the antibiotic blood level from 2.8- to 14.5-fold. Whereas the response at the lower dosage of caronamide was fairly

TABLE 2.—THE AVERAGE ANTIBIOTIC PLASMA LEVEL FOR INDIVIDUAL PATIENTS ONE HOUR FOLLOWING THE ORAL ADMINISTRATION OF PENICILLIN ALONE AND WITH CARONAMIDE

The dosage of penicillin was 100,000 units every 4 hours. The dose of caronamide was one-sixth of the total daily dosage (0.2 gm./kg./day or 0.4 gm./kg./day) administered every 4 hours with penicillin.

Patient	Penicillin control (u./cc.)	Plus 0.2 gm. caronamide (u./cc.)	Fold increase over control	Plus 0.4 gm. caronamide (u./cc.)	Fold increase over control
M. T.	0.430 (6)*	0.529 (4)	1.2	0.899 (4)	2.1
O. L.	0.282 (5)	0.321 (4)	1.1	0.600 (4)	2.1
J. Sl.	0.387 (6)	0.599 (4)	1.6	0.771 (4)	2.0
J. Su.	0.321 (6)	0.499 (4)	1.6	1.453 (4)	4.5
M. A.	0.205 (5)	0.586 (4)	2.9	0.858 (4)	4.2

* Number of samples.

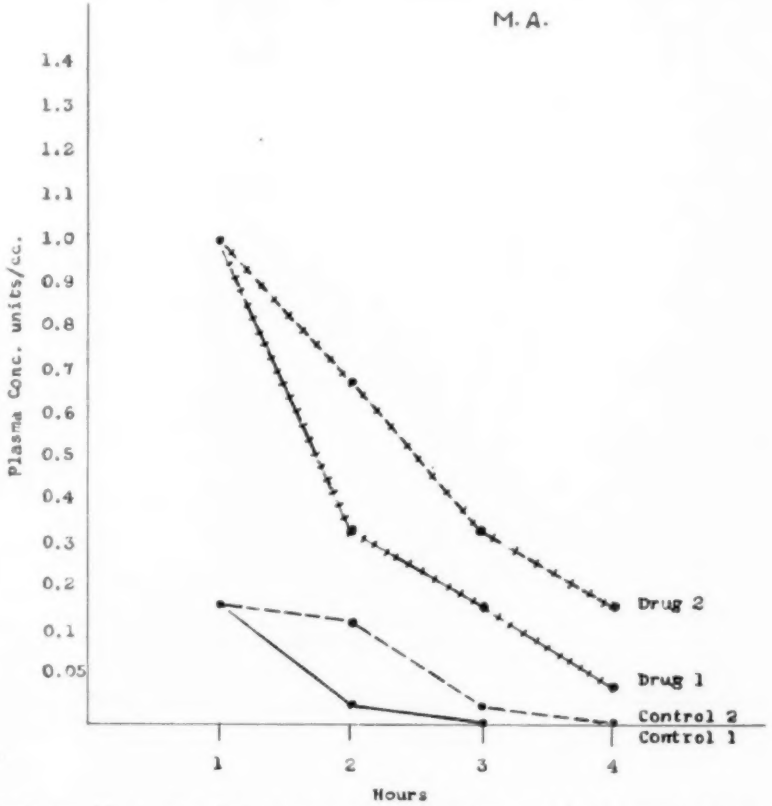

FIG. 1.—Patient M. A.: The effect of caronamide on the 4-hour antibiotic plasma level following the administration of 100,000 units of penicillin orally. C. 1 and C. 2 were two control curves wherein penicillin was administered alone. The actual dose of caronamide was given orally with penicillin 1 hour before lower drug curve was 0.5 gm. Similarly, the dose of caronamide for the top drug curve was 1 gm.

uniform at all points in the 4-hour curve there was an incommensurate increase in its effect in the latter part of the 4-hour curve at the higher dosage. It is interesting that the effect on 1-hour plasma concentrations increased fairly uniformly with the increase in caronamide dosage even though the amount of penicillin was held constant. The standard deviation of 1-hour values for the controls was ± 0.37; for the lower drug dosage, ± 0.58; and for the higher drug dosage ± 0.28.

The 1-hour values for control and drug phases for each patient have been compared in Table 2. Here the individual increase in antibiotic plasma level 1 hour following the coadministration of penicillin and caronamide was 1.1- to 2.9-fold for the 0.2 gm./kg./day dosage, and 2- to 4.5-fold for the 0.4 gm./kg./day dosage. This is probably the least favorable comparison of the data that can be made since it assumes a maximal effectiveness of the caronamide coadministration at the height of the absorption of penicillin. From such data one might deduce that ordinarily the preponderant amount of penicillin absorbed is excreted within the first hour. This would seem to be true since actually it is possible with the aid of caronamide to stop the excretion of sufficient penicillin to such an extent that its accumulation in the body amounts to as much as a 4.5-fold increase in antibiotic plasma concentration within 1 hour.

In Figures 1 to 5 are presented the

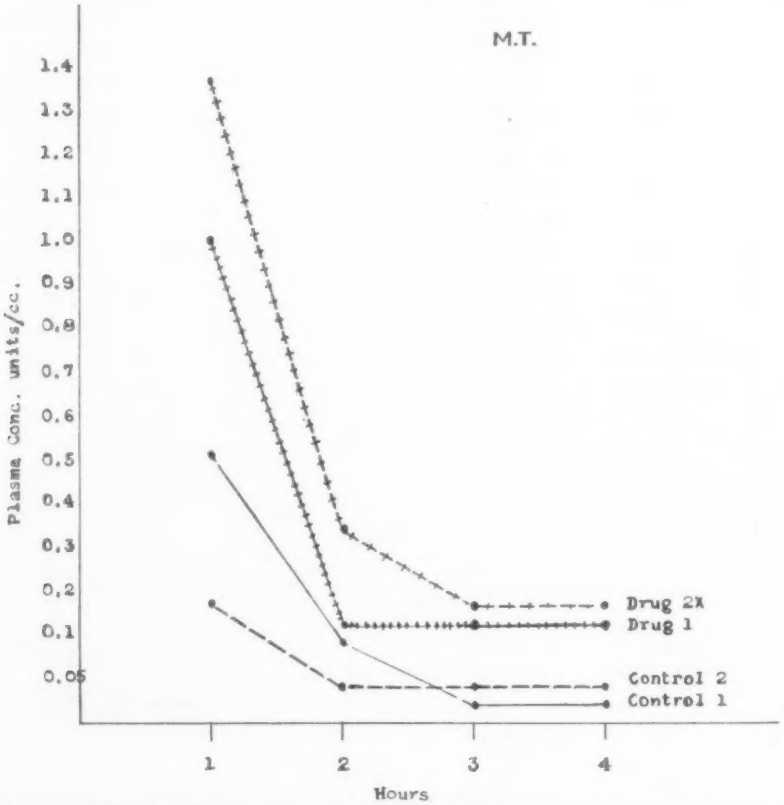

FIG. 2.—Patient M. T.: The effect of caronamide on the 4-hour antibiotic plasma level following the administration of 100,000 units of penicillin orally. C. 1 and C. 2 were two control curves wherein penicillin was administered alone. The actual doses of caronamide given orally with penicillin 1 hour before the lower drug curve was 0.5 gm. Similarly, the dose of caronamide for the top drug curve was 1 gm.

individual 4-hour dose response curves for 5 children. The curves in Figure 6 represent the average control and drug response data for the group of 5 patients. The only noteworthy difference between these data and those presented in Tables 1 and 2 is the lack of a higher initial response following the greater of the 2 caronamide dosages.

The renal function studies before and 48 hours following the eighth day of administration of caronamide are summarized in Table 3. For technical reasons some of the values in both the initial and final determinations appear "out of line." The determinations of renal function on 1 of the children who wore body casts were omitted as being unsatisfactory in both phases. It seems justifiable to conclude that caronamide did not affect irreversibly glomerular filtration, renal plasma flow, PAH Tm, or glucose Tm, even though we acknowledge the unreliability of some of the absolute values given in Table 3. The magnitude of the data presented in Table 3 is the same as Rapoport and Rubin had found previously to hold for children[9] and agrees with similar values for adults.[6] It had been ascertained previously in dogs that, whereas caronamide did decrease the clearance of PAH when the two were administered together, it did not affect glomerular filtration, urea clearance, glucose Tm, arginine Tm, or sulfonamide clearance.[1]

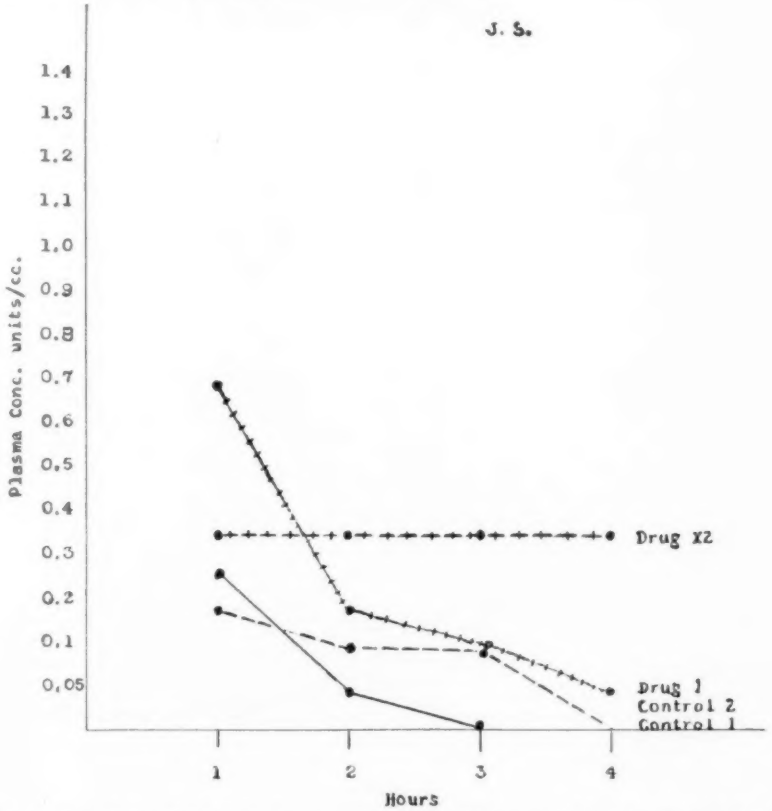

FIG. 3.—Patient J. S.: The effect of caronamide on the 4-hour antibiotic plasma level following the administration of 100,000 units of penicillin orally. C. 1 and C. 2 were two control curves wherein penicillin was administered alone. The actual dose of caronamide given orally with penicillin 1 hour before the lower drug curve was 0.5 gm. Similarly the dose of caronamide for the top drug curve was 1 gm.

TABLE 3.—RENAL FUNCTION STUDIES BEFORE AND FOLLOWING THE ORAL ADMINISTRATION OF CARONAMIDE IN A DOSAGE OF 0.2 TO 0.4 GM./KG./DAY FOR A PERIOD OF 8 DAYS

These values have been corrected to a body surface area of 1.73 sq. m.

	Patients			
	O. L.	M. T.	J. Su.	M. A.
Surface area, sq. m.	0.98	0.85	0.88	0.71
Glomerular filtration rate, cc./min.:				
B*	125	158	111.8	136
A†	160	126.8	94.8	127
Renal plasma flow, cc./min.:				
B	810.7	802	527	609
A	732.8	732	533	638
PAH Tm, mg./min.:				
B	97.9	112	33.5	50.4
A	74.7	76.1	55.4	76
Glucose Tm, mg./ min.:				
B	341	288	292	376
A	478	241	329	409

* B = Before therapy (control).
† A = After therapy (drug effect).

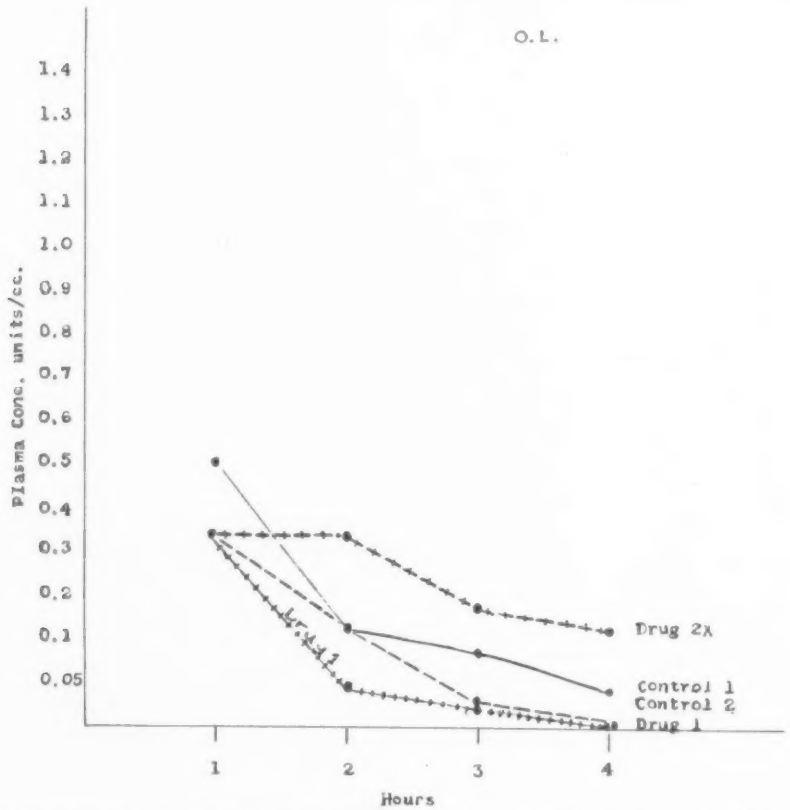

FIG. 4.—Patient O. L.: The effect of caronamide on the 4-hour antibiotic plasma level following the administration of 100,000 units of penicillin orally. C. 1 and C. 2 were two control curves wherein penicillin was administered alone. The actual dose of caronamide given orally with penicillin 1 hour before the lower drug curve was 0.5 gm. Similarly, the dose of caronamide for the top drug curve was 1 gm.

Summary. When tested in children, the oral administration of caronamide at a dosage of 0.2 gm./kg./day together with penicillin increased the antibiotic plasma concentration 1.8- to 2.8-fold over the control values when penicillin was administered alone. When the dosage of caronamide was increased to 0.4 gm./kg./day the penicillin plasma concentration was increased 2.8- to 14.5-fold over the control figures.

There was no instance of toxicity in these patients that warranted discontinuance of caronamide when the compound was administered in oral dosages of 0.1 to 0.4 gm./kg./day over a period of 1 or 2 weeks. Although there were 2 urine specimens that contained drug crystals and 3 instances of rash, in which no 2 cases were similar in appearance or distribution, these did not require special attention. Two of the rashes cleared up during continued administration of drug and the third appeared 24 hours after the drug had ceased to be given. There were 3 cases in which one or more isolated instances of "pentosuria" were found, and these were associated with a pharyngitis, smallpox vaccination, or the rash that appeared after the drug had been stopped.

By means of renal function tests run before and at the end of the coadministration of penicillin and caronamide it was ascertained that the latter compound did not influence irreversibly glomerular filtration, renal plasma flow, PAH Tm, or glucose Tm.

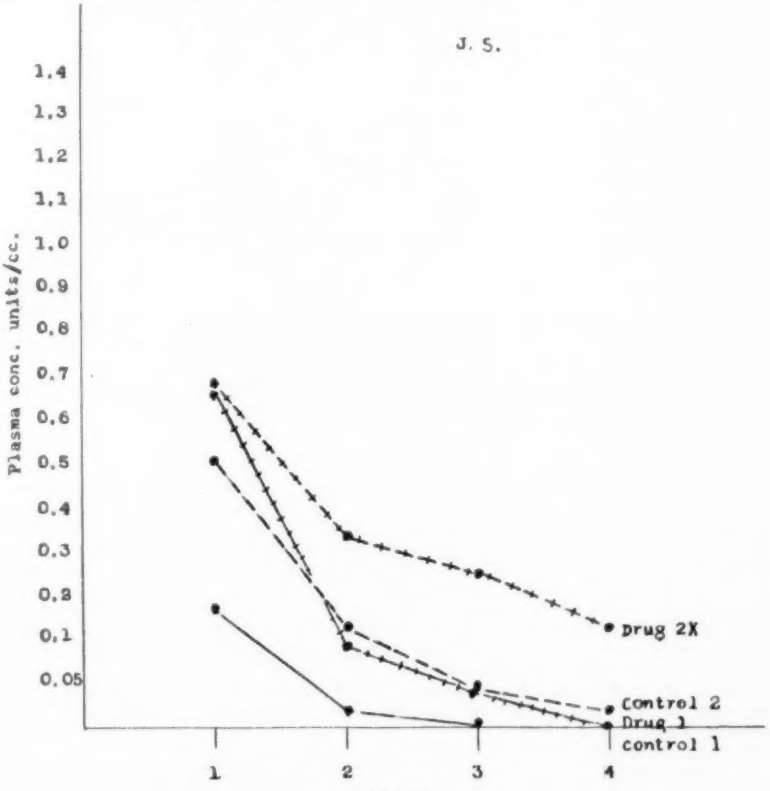

FIG. 5.—Patient J. S.: The effect of caronamide on the 4-hour antibiotic plasma level following the administration of 100,000 units of penicillin orally. C. 1 and C. 2 were two control curves wherein penicillin was administered alone. The actual dose of caronamide given orally with penicillin 1 hour before the lower drug curve was 0.5 gm. Similarly, the dose of caronamide for the top drug curve was 1 gm.

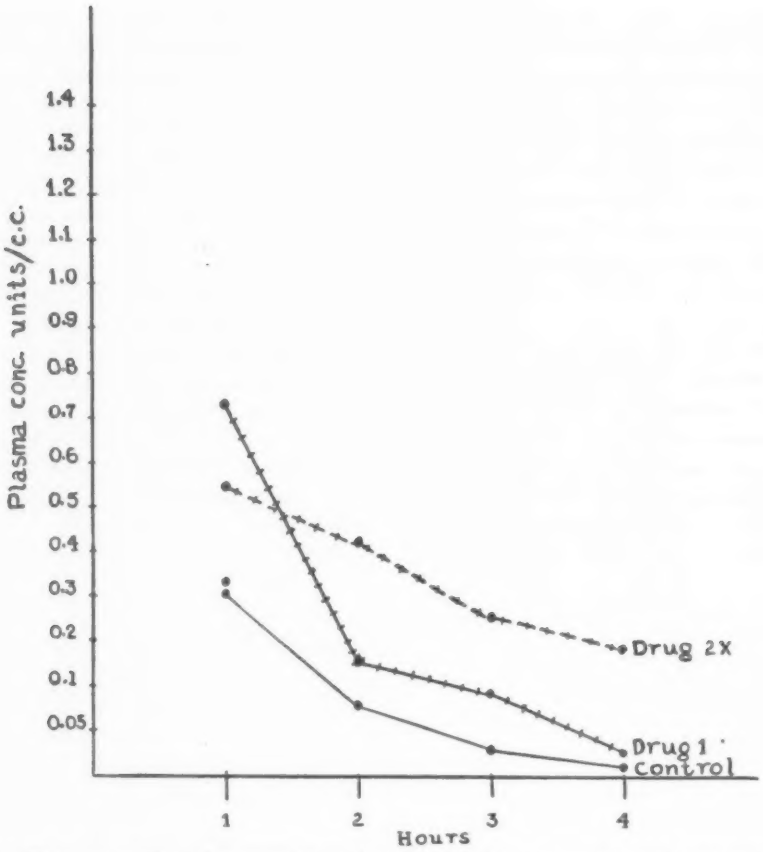

FIG. 6.—The average effect of caronamide on the 4-hour antibiotic plasma level following the administration of 100,000 units of penicillin orally. The control curve represents an average of all the corresponding values for both control phases. The coördinate for the lower drug curve are averages of all the corresponding penicillin values when the dose of caronamide administered with each dose of penicillin was one-sixth the daily dose of 0.2 gm./kg. Similarly, the caronamide dose for the top drug curve was one-sixth the daily dosage of 0.4 gm./kg.

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MYELOPHTHISIC ANEMIA THE PRESENTING MANIFESTATION OF PROSTATIC CARCINOMA WITH SKELETAL METASTASES: THE EFFECT OF CASTRATION AND STILBESTROL

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Carcinoma of the prostate, although frequently metastatic to bone, is not often associated with severe anemia. It is the purpose of this communication to point out that myelophthisic anemia may be the presenting manifestation of prostatic cancer and that this anemia may respond to combined castration and female sex hormone administration.

Case Reports. Case 1. J. W., a married painter of 47, was admitted to the Boston City Hospital August 17, 1929, complaining of pallor, weakness, loss of 75 pounds in weight, and pains in the arms and legs, all with onset approximately 9 months earlier. The pains had at first been intermittent but later were described as being almost continuous and extending throughout the length of all four limbs. On occasions the pains in the arms had been shooting in character. Five months before admission he had suffered from acute urinary retention which required catheterization. There had been no other urinary symptoms prior to this nor did any develop subsequent to this single episode. Eight weeks before admission the patient awakened one morning with a left facial paralysis. He stated that on occasions he had noted edema of the legs during his illness. His past and family histories were irrelevant. He denied venereal disease.

Examination revealed an extremely pale, markedly emaciated, white man in no apparent distress, but appearing much older than the stated age. The peripheral arteries were markedly thickened. There was generalized weakness and atrophy of all the muscles of

Table 1.—Blood Findings Upon Admission in 3 Cases of Myelophthisic Anemia Secondary to Carcinoma of the Prostate With Skeletal Metastases

	Case 1	Case 2	Case 3
Red blood cells (millions per cmm.)	2.15	2.74	2.30
Hemoglobin (gm. per 100 cc.)	5.6	7.2	6.8
White blood cells (thousands per cmm.)	3.4	5.0	7.0
Hematocrit (%)	..	24.4	24.0
Mean corpuscular volume (c. micra)	..	89.0	104.2
Mean corpuscular hemoglobin concentration (%)	..	33.9	34.3
Reticulocytes (%)	4.8	4.0	2.0
Nucleated red blood cells	+	0	+
Anisocytosis	+++	+	+++
Poikilocytosis	+++	+++	+++
Platelets	128,000	Increased	Increased
Icteric index	4	3	5
Non-protein nitrogen (mg. per 100 cc.)	42	27	37
Differential:			
Polymorphonuclear neutrophils (%)	54	67	70*
Eosinophils (%)	..	2	4
Basophils (%)	1	1	..
Lymphocytes (%)	38	11	18
Monocytes (%)	7	19	8

* Bands 29.

the trunk and limbs, and pressure over muscles revealed moderate tenderness. Neurologic examination was essentially negative except for a peripheral type of left facial palsy. Heart, lungs, abdomen and skeleton were normal on physical examination. The prostate was of normal size, firm and smooth. Laboratory examinations showed the specific gravity of the urine to range from 1.005 to 1.010 with albumin and white cells constantly present. The blood non-protein nitrogen was 42 mg. per 100 cc. No free hydrochloric acid was present in the gastric juice after the subcutaneous injection of histamine. Lumbar puncture was negative. Stools were negative for occult blood. Roentgen ray examination of the skeleton revealed widespread metastatic involvement of an osteoplastic type such as commonly originates in the prostate. Although most marked in the trunk, hip and shoulder girdles, and proximal portions of all four extremities, lesions could be seen extend-

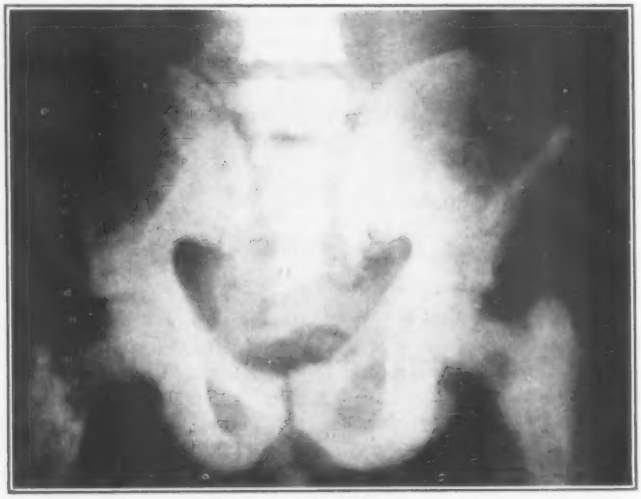

FIG. 1.—(Case 1.) Roentgen ray of pelvis.

FIG. 2.—(Case 1.) Roentgen ray of feet.

ing to the distal tips of the fingers and toes (Figs. 1 and 2). The results of blood examination are shown in the table.

Transfusion of 500 cc. of whole blood was performed and a trial course of iron therapy was begun, which was not followed by any change in blood values. Two hundred grams of whole liver daily followed by 30 gm. of liver extract (Lilly 343) daily was unassociated with any response. A second transfusion of whole blood was given 3 weeks after admission. On October 1 gross hematuria occurred for 12 hours and then disappeared. Following this episode the patient had some difficulty in commencing urination and occasionally was incontinent. At this time repeated examination of the prostate showed it to be fixed and very firm although entirely smooth. The patient's anemia became increasingly severe and in November the prostate felt stony hard and nodular. On November 19, approximately 1 year after the onset of symptoms, the patient died. Postmortem examination was limited to the prostate which although stony hard appeared grossly normal. Microscopic examination revealed scirrhous carcinoma.

Case 2. H. S., a married caretaker, 64 years of age, was admitted to the Boston City Hospital, December 1, 1933, complaining of increasing weakness, pallor, and loss of weight, with onset 6 months previously. For a few weeks there had been some aching in the shoulders, and in the muscles of the legs. For 3 weeks he had received iron and liver extract treatment with no noticeable effect upon his condition. There had been no urinary symptoms of any kind. His family and past histories were irrelevant. Venereal disease was denied.

Examination revealed a thin, pale, white man lying comfortably in bed. All but 3 teeth were absent. These were carious and surrounded by pyorrhea. There was moderate tenderness of the calf and shoulder muscles on pressure and atrophy and lack of tone of the calf muscles. Neurologic examination was negative. The remainder of the physical examination was normal. Rectal examination revealed a small, smooth, fixed, stony hard prostate. The urine was straw, alkaline, specific gravity 1.014, albumin negative, sugar negative, microscopic negative. Examination of the blood is shown in the table. Roentgen rays of the chest and shoulders were negative. Roentgen rays of the pelvis and spine showed extensive osteoplastic metastases involving the entire pelvis and the lumbosacral spine. The patient was discharged to the care of his private physician. He died at home several months later.

Case 3. H. G., a single retired barber, 78 years of age, was admitted to Cushing Veterans Administration Hospital on November 30, 1946, complaining of increasing weakness, fatigability and exertional dyspnea of 1 year's duration. Three months before admission he developed constant pain in the left axillary line at the level of the 7th rib. For several months he had nocturia and slight difficulty in starting his urinary stream, with dribbling. His family history was negative. His past history revealed recurrent malaria for several years subsequent to his initial infection in Cuba in 1898. "Creaking" of both shoulder joints on motion had been present for 20 years and occasional transitory chest and low back pain for 7 years.

Examination revealed a pale, thin, edentulous, white man in no apparent distress. The anterior-posterior chest diameter was moderately increased. The percussion note was hyperresonant. Expiratory breath sounds were prolonged. A non-tender, smooth liver edge was felt 3 cm. below the costal margin. The upper border of liver dullness was in the 8th interspace by percussion. Skeletal examination was negative except for crepitus upon movement of the shoulder joints. The prostate was enlarged, fixed, hard and contained several stony nodules. Laboratory examinations showed the urine specific gravity to vary from 1.008 to 1.020. Albumin and sugar were absent. Rare white blood cells and occasional red blood cells, hyaline and granular casts were present. Stools were negative for occult blood. Kahn test negative. The blood nonprotein nitrogen was 37 mg., serum calcium 9.12 mg., phosphorus 4.3 mg., total plasma proteins 6.3 gm., albumin 3.3 gm., and globulin 3 gm. per 100 cc. Alkaline phosphatase—16.8 Bodansky units. Roentgenograms of the entire skeleton showed widespread osteoplastic metastases throughout the pelvis, upper ends of the femora, thoracic and lumbar spine and ribs.

Ferrous sulfate 0.33 gm. 3 times daily by

mouth and 15 units of concentrated liver extract daily by intramuscular injection were administered for 3 weeks. No rise in reticulocytes occurred and there was a decline in red blood cells to 2,000,000 per cu. mm. and in hemoglobin to 6.2 gm. per 100 cc. During the 4th week the patient was castrated and 5 mg. of diethylstilbestrol was given by mouth daily. Blood studies immediately after operation showed the red blood cells to be 1,990,000 per cu. mm. and the hemoglobin 5.1 gm. per 100 cc. The hematocrit was 21%. Reticulocytes were followed for several weeks. No increase occurred. A gradual rise took place in red blood cells, hemoglobin and hematocrit to values of 3,500,000 per cu. mm., 11 gm. per 100 cc. and 34% 2 months after castration. Concurrently, nucleated red blood cells disappeared from the peripheral blood and the number of band form polymorphonuclear leukocytes decreased to less than 5%. The patient's strength and sense of well-being improved markedly. Bone pains disappeared. This clinical improvement was maintained. The blood levels on July 18, 1947, 7 months after castration, were: Red blood cells, 4,000,000 per cu. mm., hemoglobin 12 gm. per 100 cc., white blood cells 6500 per cu. mm., hematocrit 36%, mean corpuscular volume 90 cu. micra; polymorphonuclear neutrophils 74% (band forms 1%), lymphocytes 25%, monocytes 1%. The red blood cells appeared normal on stained films; there was no polychromatophilia and no nucleated erythrocytes were present. Moderate regressive changes were noted in the roentgenologic appearance of the skeletal metastases. The blood values continued to remain at the same level during the next 6 months. The last observation was made Jan. 24, 1948. The patient continues asypmtomatic 13 months after the institution of therapy.

Discussion. The above cases illustrate the fact that severe myelophthisic anemia accompanied by weakness and loss of weight may be the presenting manifestations of carcinoma of the prostate with extensive skeletal metastases. Examination of the prostate in Case 1 revealed little in the way of abnormality at the time of admission. Later is this case, and in the second case on admission hardness and fixation of the prostate were found. However, without the Roentgen ray evidence of skeletal involvement the diagnosis of carcinoma of the prostate would have been more difficult to establish.

In Case 1 the existence of metastatic lesions extending to the distal phalanges of the hands and feet is rather remarkable since metastatic lesions rarely are found below the knee or elbow. The mechanism of myelophthisic anemia remains somewhat obscure although it would appear reasonable to believe that the hematopoietic marrow has been crowded out by the abnormal tissue. Vaughan[14] believes that certain cancers deprive the hematopoietic cells of necessary nutritional factors, thus leading to an imperfect formation of blood cells.

Several recent reports have noted improvement in anemia coincidental with the modern treatment of prostatic carcinoma but no blood values have been reported.[6,7]

A question which naturally arises is whether the effect of castration and estrogens in the last case was directly upon blood formation or indirectly through an effect on the cancerous metastases. The absence of testicular function, as in eunuchs, tends to lead to slight anemia. In 7 out of 8 such cases given methyl testosterone or testosterone propionate or both, the red blood cells and hemoglobin increase.[10] Castration of rats leads to a moderate decline in red blood cell and hemoglobin values,[4] and regeneration of blood after induced hemorrhage in such animals is less rapid than in normal rats.[5] The administration of androgens to castrated rats leads to marrow hyperplasia and red blood cell increases,[4] and similarly speeds the regeneration of red cells following induced hemorrhage in both normal and castrated rats.[5] Androgen administration in rats prevents the pancytopenia which follows hypophysectomy.[4] There is thus little evidence which would suggest that castration with the concomitant decline in androgen production would directly

lead to an increase in red blood cell formation; rather the reverse might be anticipated.

In therapeutic doses in women there is no evidence that stilbestrol produces any change in the blood.[8,9,15] In rats no change in the bone marrow or the peripheral blood occurs with doses comparable to those used in man.[12] With excessive doses, ranging from 5 to 500 times the usual maximum therapeutic dose, rats develop anemia, leukopenia, thrombocytopenia and bone marrow hyperplasia followed by hypoplasia.[9,11,12] In dogs massive doses first produce leukocytosis which is rapidly followed by leukopenia (eventually culminating in agranulocytosis), anemia, thrombocytopenia and bone marrow hypoplasia.[1,2,3,9,13] The administration of 10 mg. of diethylstilbestrol to adult rhesus monkeys has been reported as having no effect on the blood except for slight anemia,[3] but following the administration of carbon tetrachloride a similar quantity leads to marked anemia.[3]

Similar effects to those obtained with stilbestrol have occurred following the administration of estradiol benzoate to rats[9] and to dogs.[2,3,13] Estradiol has likewise been shown to retard red blood cell and hemoglobin regeneration after induced hemorrhage in rats.[5] There is thus no evidence that stilbestrol stimulates erythropoiesis, but rather that in large doses it produces pancytopenia with bone marrow hypoplasia.

Although much of the above work was performed on animals and thus may not be strictly applicable to man, the fact that in Case 3 nucleated erythrocytes disappeared from the peripheral blood and the number of band form polymorphonuclear neutrophils decreased markedly suggests that either one or both of the therapeutic procedures directly affected the carcinoma in the bone marrow. Whether this effect is a specific one for prostatic carcinoma or whether the effect is obtained by virtue of an action of estrogens on the bone producing or destroying cells is not clear. It is felt that the results obtained warrant a trial of estrogenic therapy in cases of myelophthisic anemia due to metastases from other types of carcinoma.

Summary. Three patients with myelophthisic anemia as the presenting manifestation of carcinoma of the prostate with widespread skeletal metastases have been presented. The first two of these patients were observed before present-day methods of treating carcinoma of the prostate were evolved. Each died within a few months of diagnosis with anemia controlled only by blood transfusion. The third patient, whose blood values declined during a preliminary period of treatment with iron and liver extract, was castrated and subsequently treated with 5 mg. of diethylstilbestrol daily, following which the blood values returned nearly to normal. Symptoms referable to skeletal involvement disappeared. This improvement was still present 7 months later. Although the mechanism of this response is unclear, it is suggested that the treatment so affected the metastatic cancerous tissue invading the bone marrow that more normal hematopoiesis was able to take place.

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ANTIHISTAMINIC, ANTIANAPHYLACTIC AND ANTIALLERGIC ACTIVITY OF N-(a-PYRIDYL)-N-(a-THENYL)-N′, N′-DIMETHYL-ETHYLENEDIAMINE HYDROCHLORIDE*

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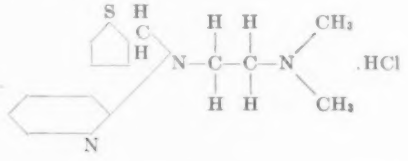
Interest in the field of antihistamine therapy has stimulated a search for newer compounds possessing greater activity in allergic states than those presently available, and which at the same time may prove to be less toxic when used clinically. A promising development in this line of endeavor has been the synthesis[6] of N-(a-pyridil)-N-(a-thenyl)-N′, N′-dimethylethylenediamine hydrochloride, supplied to us for this study under the name of Thenylene.*



This compound is closely related to Pyribenzamine and Neoantergan and differs in chemical structure by the substitution of a thiophene radical for the benzyl or methoxybenzyl ring. Thenylene, in common with other antihistaminics previously investigated, possesses marked specificity against the pharmacologic actions of histamine. It abolishes the histamine contraction of the isolated guinea pig ileum, markedly reduces the vasodepression induced by histamine in the anesthetized cat, and protects the guinea pig against the lethal effects of aërosolized or intravenous histamine. Activity against the acetylcholine and barium chloride contractions of the rabbit's intestine is relatively small as compared to atropine and papaverine.[5] Acute toxicity in mice[3] is comparable to Pyribenzamine and Benadryl. Animals receiving 20 mg. per kg. of the drug for 4 weeks showed no pathologic organ changes. There have been noted in other studies, pathologic changes in the liver of some animals receiving massive doses over prolonged periods of time.[4]

In the present investigation, the antihistaminic, antianaphylactic, and antiallergic properties of Thenylene were studied in order to make some comparison with other drugs of this series, and to gain some insight, if possible, into the relationship of antihistaminic and antianaphylactic activity to clinical efficacy in allergic states.

Antihistaminic Activity. A standard protective dose of 3 mg. per kg. of Thenylene was administered intraperitoneally to a group of male guinea pigs, 400 to 500 gm. in weight, 15 minutes prior to the intravenous injection of histamine (Table 1).

Table 1.—Protective Action of Thenylene, 3 Mg. per Kg., Against Increasing Amounts of Histamine

Histamine I.V. (mg. (base)/kg.)	No. animals	Mortality (%)
0.4	6	0
0.8	8	13
0.12	7	29
2.0	8	50
3.0	10	50
4.0	10	80
5.0	10	90
6.0	10	100

* The experimental work in this study was supported by a grant from Abbott Laboratories, North Chicago, Illinois.

* Thenylene brand of N-(a-pyridyl)-N-(a-thenyl)-N′, N′-dimethylethylenediamine hydrochloride was supplied by the Abbott Research Laboratories, North Chicago, Illinois. The same drug is undergoing clinical trial by Eli Lilly & Co., under the designation of Histadyl.

The 100% lethal dose of histamine when injected into the penile vein of male guinea pigs has been determined to be 0.4 mg. per kg. of the base.[1] This standard dose of Thenylene protected all animals against 1 lethal dose of histamine, while 50% of the animals survived as much as 3 mg. per kg. of histamine, approximately 8 lethal doses. Six mg. per kg. of histamine, or 15 lethal doses, produced 100% mortality in this group of Thenylene-treated animals. The protective action of decreasing amounts of Thenylene against 1 lethal dose of histamine was also determined (see Table 2). One mg. per kg. of Thenylene

TABLE 2.—PROTECTIVE ACTION OF DECREASING AMOUNTS OF THENYLENE AGAINST 1 LETHAL DOSE (0.4 MG./KG. BASE) OF HISTAMINE

Thenylene (mg./kg.)	No. animals	Mortality (%)
1.0	10	0
0.1	10	60
0.01	10	100

prevented death in all animals tested, while 0.1 mg. per kg. protected 4 out of 10 guinea pigs against fatal shock. A dose of 0.01 mg. per kg. afforded no protection.

ANTIANAPHYLACTIC ACTIVITY. Male guinea pigs, 400 to 500 gm. in weight, were sensitized by the intraperitoneal injection of 0.1 cc. of normal horse serum. Twelve days later a shocking dose of 1 cc. of the same serum was administered intravenously (see Table 3). Of the controls,

TABLE 3.—PROTECTIVE ACTION OF DECREASING AMOUNTS OF THENYLENE AGAINST ANAPHYLACTIC SHOCK

Thenylene (mg./kg.)	No. animals	Mortality (%)
0 (control)	14	100
1.0	10	20
0.1	14	43
0.01	10	100

100% died in typical anaphylactic shock. The remaining guinea pigs were divided into 3 groups, and given 1,0.1 and 0.01 mg. per kg. of Thenylene respectively by intraperitoneal injection, 15 minutes prior to the shocking dose of antigen. Fatal anaphylaxis occurred in 20% of the animals receiving 1 mg. per kg. of Thenylene. A mortality of 43% occurred in the group pretreated with 0.1 mg. per kg. Those receiving 0.01 mg. per kg. were not protected against fatal anaphylactic shock.

ANTIALLERGIC ACTIVITY. The clinical action of Thenylene was studied in 117 patients with 1 or more allergic complaints (see Table 4). Twenty patients had 2

TABLE 4.—CLINICAL RESULTS WITH THENYLENE

	No. treated	Improved	Unimproved
Urticaria:			
Acute	5	4	1
Chronic	9	7	2
Seasonal hay fever	40	30	10
Rhinitis, perennial allergic	50	32	18
Asthma	21	7	14
Dermatitis:			
Atopic	2	1	1
Contact	5	4	1
Unclassified	2	0	2
Headache, allergic	2	0	2
Pruritis ani, allergic	1	0	1

complaints, and the effect on each was not necessarily the same. A favorable effect on certain allergic symptoms was observed, similar in nature to that exerted by other known antihistaminics. Clinical effect when present, occurred within 30 minutes following oral administration, and lasted several hours. The drug was usually administered to adults in doses of 100 mg., while one-quarter to one-half this amount was given to children. In those with constant symptoms, the drug was prescribed 4 times daily, after meals and at bedtime. Where symptoms were intermittent it was recommended only when necessary. In many instances it was found that smaller amounts than originally prescribed were adequate in controlling symptoms.

The majority of patients with urticarial dermatoses noted a reduction of lesions and pruritus following each dose of drug. In 5 cases of acute urticaria, all but 1 experienced marked symptomatic relief, while 7 of 9 patients with chronic urticaria noted a favorable effect. Approximately 75% of acute seasonal hay fever obtained some benefit from the drug, although relief of symptoms was seldom complete. The mild discomfort occurring in some

patients who had received protective immunization against the specific allergen to which they were sensitive appeared to be more greatly benefitted than the severer symptoms of unimmunized individuals. In many cases the drug appeared to be more effective in alleviating symptoms at the onset and height of the ragweed pollinating season than in the latter days when the pollen count was declining. This has also been observed to be the case with other antihistaminic drugs. Symptomatic benefit was somewhat less in cases of non-seasonal allergic rhinitis, 64% of patients noting some improvement. The drug was used in 21 cases of asthma with benefit occurring in 7 patients. Relief of symptoms in the latter was not striking, and usually not as great as that obtained from older remedies such as epinephrine, ephedrine or aminophylline. The pruritus of atopic and contact dermatitis was notably diminished in 5 of 7 patients. In 2 other cases where the type of dermatitis could not be accurately classified, but which were felt to be allergic in nature, no help was evident. Results were negative in 2 instances of allergic headache, and in 1 case of pruritus ani.

Side Action. One or more side effects, usually mild in nature, were encountered in approximately 25% of the patients receiving the drug. These occurred most often when doses of 100 mg. were administered but rarely affected the continued administration of the drug. A reduction in dosage to 50 mg. obviated the side action in most instances. Drowsiness, which is the most common side effect noted with other antihistaminics, occurred in 13 patients. Vertigo, headache, gastro-intestinal distress and excessive dryness of mucous membranes were next in order of frequency. No serious toxic action was observed in any patients in this group. Few received the drug continuously for periods of more than 2 weeks. In 1 patient who received 400 mg. daily for 10 weeks, and in 2 others who received the same dosage for 8 weeks, no changes in the peripheral blood or urine were observed. In view of the liver changes occurring in laboratory animals receiving large doses over prolonged periods[4], it would seem advisable to follow closely with laboratory studies any patient receiving large amounts of the drug for a long period of time.

Discussion. Thenylene compares favorably with other antihistaminic agents already in use in this country and abroad. Its activity against histamine is somewhat less than that of Pyribenzamine or Neoantergan when compared on the basis of protection afforded the guinea pig against multiple lethal doses of histamine.[2] When its effectiveness is measured against 1 lethal dose of histamine, or by its ability to prevent fatal anaphylaxis, it is closely comparable to other active drugs of this series. While one is tempted to make clinical comparisons between symptomatic drugs in allergy, this must be done with due regard to the variability of the allergic state in the same individual at different times. Thenylene, in the dosage employed in this study, exerts a palliative action in many allergic conditions of a similar order shown by other antihistaminics currently available. As more drugs of this group are synthesized, it becomes evident that patients who fail to obtain a favorable action from 1 compound, may react more satisfactorily to a related compound. The relatively low incidence of side action at effective dosage entitles Thenylene to a place in the growing list of palliative agents in allergy. These drugs, of course, do not replace etiologic investigation, the elimination of offending allergens, or desensitization where indicated, but act as helpful adjuncts in the control of the allergic state.

Summary. 1. N - (a - pyridyl) - N - (a - thenyl) - N′, N′ - dimethylethylenediamine hydrochloride (Thenylene) was investigated with reference to its effectiveness in preventing fatal histamine shock and anaphylaxis in guinea pigs, and in alleviating allergic symptoms in man.

2. As many as 15 lethal doses of histamine were required to kill some animals protected with 3 mg. per kg. of Thenylene. One mg. per kg. of the drug protected all animals against 1 lethal dose of histamine, while 0.1 mg. per kg. protected 40% against this fatal dose. Marked protection against fatal anaphylaxis in guinea pigs was observed with as little as 0.1 mg. per kg. of Thenylene.

3. Symptomatic relief for several hours following each dose of the drug was evident in many cases of urticaria, hay fever and perennial allergic rhinitis. Results in asthma were not striking. The pruritus of allergic dermatoses were alleviated in some instances.

4. Mild side effects, such as drowsiness, vertigo and gastro-intestinal upset, occurred in 25% of those receiving the drug. These were rarely severe enough to warrant discontinuation of the drug.

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PERFORATION OF THE INFARCTED INTERVENTRICULAR SEPTUM

REPORT OF TWO CASES, ONE DIAGNOSED ANTEMORTEM

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PERFORATION of the interventricular septum as a complication of cardiac infarction is a syndrome occasionally found at autopsy and rarely diagnosed before death. The purpose of this article is to review the literature on this subject and to add 2 more cases to those previously reported. One of the present cases is the fifteenth reported case in which the diagnosis was made antemortem.

In 1934 Sager[13] reviewed the literature with regard to perforation of the infarcted interventricular septum and reported a case of his own. At that time, including Sager's case, a total of 18 cases had been reported. Until then only 1 case had been diagnosed antemortem, Sager's case being the second. In 1943, Weber[15] again reviewed the literature of this subject and reported a case of his own. Sixteen additional cases of this entity were found, of which 5 were diagnosed antemortem. Since Weber's review, 7 additional cases had been reported through July 1946 in addition to 13 cases found by Edmondson and Hoxie.[5] Seven of these 20 cases were diagnosed antemortem. Thus, a total of 54 cases of this condition had been reported through July 1946, of which 14 were diagnosed antemortem. Weber's[15] patient was a 46 year old man, who was seen 9 days after the onset of precordial pain with a rough systolic murmur over the entire precordium, maximal in the fourth and fifth left intercostal spaces and a palpable thrill in the latter area. He died 20 days after the onset of pain. Lober and Hertzog[9] reported the case of a 68 year old white woman who developed precordial pain 7 days prior to examination. When first observed she had a systolic thrill and murmur in third and fourth interspaces to the left of the sternum. She died the day of admission. Master and Russell[11] report a 47 year old man who had a typical history for myocardial infarction and ECG changes of an anterior myocardial infarction. On the 3rd day of his illness, he developed a systolic thrill and systolic murmur in the fourth left interspace. Right axis deviation developed on ECG and Q3 appeared. The patient died on the 11th day of his illness. At autopsy there was acute infarction at the apex of the left ventricle as well as 2 perforations of the interventricular septum.

Wood and Livezey[17] report an unusual case who survived 4 years and 10 months after his acute episode of septal perforation. The patient was a 44 year old white man seen 1 month and 6 days after coronary pain appeared. He had a loud rough systolic murmur and systolic thrill over the lower sternum. Two weeks later he developed right heart failure and 6 weeks later dependent edema which never left him. He was benefited by mercurials but very little by digitalis. At necropsy the right coronary artery was thrombosed and there was a 1.5 cm. perforation in the interventricular septum. Edmondson and Hoxie,[5] in a review of 25,000 consecutive necropsies performed at the Los Angeles County Hospital over a 17 year period, found 865 unhealed infarcts, 72 of which

had ruptured, and 13 of which had ruptured interventricular septa. Three of these were diagnosed antemortem. The time of survival was known in 11 cases; the average of 10 was 2.25 days, the other surviving several months. In 11 of the 13 a systolic murmur was observed. Benson, Hunter and Manlove,[3] reviewing 7000 autopsies, found 40 cases of cardiac rupture, 2 of which had perforated septa. One of these 2 was a 60 year old man found dead in bed; he had not been previously examined. The second was an 81 year old man who became dyspneic 8 days prior to admission. On admission he had a loud apical systolic murmur transmitted to the axilla and back. He died on the 2nd hospital day. Wood[16] reported a case in a 69 year old white woman who had a severe attack of precordial pain 10 days before admission. There was on admission a blowing systolic murmur over the lower sternum, transmitted to the axilla and up the sternum. There was no thrill. ECG showed evidence of recent posterior infarction. The patient expired 5 weeks after admission.

Foster[6] reports the case of a 67 year old white man who was hospitalized soon after the onset of substernal pain with radiation down the left arm. Physical findings are not given in his report. At autopsy, 3 days later, there was found an anterior myocardial infarct with septal involvement and a septal perforation 1 cm. in diameter located anteriorly near the cardiac apex.

Bean[2] in his review of 300 cases of cardiac infarction collected from the records of the Boston City Hospital found 1 instance of infarcted septum with fresh perforation. Through the courtesy of this author we are able to report this case in more detail than appeared in his original article.

Case Reports. Case 1. A 35 year old white American woman was admitted to the Boston City Hospital on Jan. 17, 1936. Her past history was negative except for known diabetes. The history was suggestive of anginal pain first occurring 7 days before hospital admission.

P. E. P. 100; R. 30; B.P. 0/0. Obese white woman in coma and shock on admission. Heart percussed normal in size. Heart sounds were clear. There were no murmurs. Kussmaul breaking was noted. The lungs were clear to percussion and auscultation.

Laboratory Data. WBC 7000. Urine showed albumin, sugar, casts. Blood sugar 480 mg. per 100 cc. NPN 80 mg. per 100 cc.

Course. Patient was given insulin, fluids but no digitalis. She expired during the 1st hospital day.

Necropsy Findings. The heart weighed 320 gm. with body weight of 70 kg. There was marked sclerosis of the left coronary artery. The heart valves were normal. There was a fresh thrombus in the left anterior descending coronary artery occluding this vessel. The anterior portion of the interventricular septum and adjacent portions of the right and left ventricular walls were infarcted. In the anterior portion of the septum near the apex was a ragged, irregular perforation 1.5 cm. in diameter. The lungs showed moderate congestion and edema. Liver, kidneys and spleen were not remarkable. There was generalized arteriosclerosis, no evidence of emboli or other thrombi.

Case 2. L. H., a 76 year old white woman, was admitted to the medical service of the Cincinnati General Hospital on July 10, 1947. Two weeks prior to admission the patient developed a productive cough with some substernal chest pain on coughing. She also complained of pain in the abdomen, back, left hip and left leg. For a week prior to admission she had been febrile, had had chilly sensations, and had been confined to bed. During this week she had been semiconscious most of the time with occasional lucid intervals; also during this period she vomited frequently, and her breathing was observed to be labored. She became completely unresponsive on the day of admission.

P. E. T. 101; P. 64; R. 48; B.P. 120/80. The patient was a moderately obese, dyspneic, slightly cyanotic white female. She was not orthopneic. She was disoriented but responded to commands in a confused manner. Neck: no venous distention.

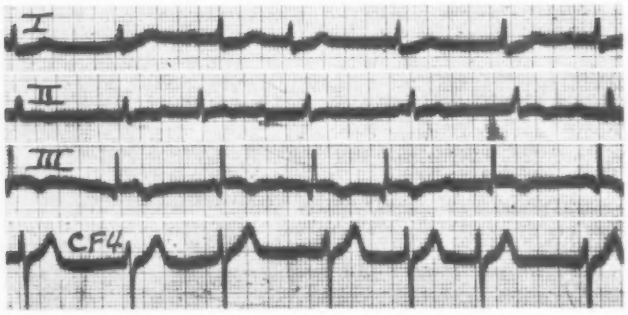

Fig. 1.—Case 2. ECG (July 10, 1947) showing Q_3 and elevated ST_3 consistent with posterior left ventricular infarction. Reciprocal ST depression in leads I and CF_4.

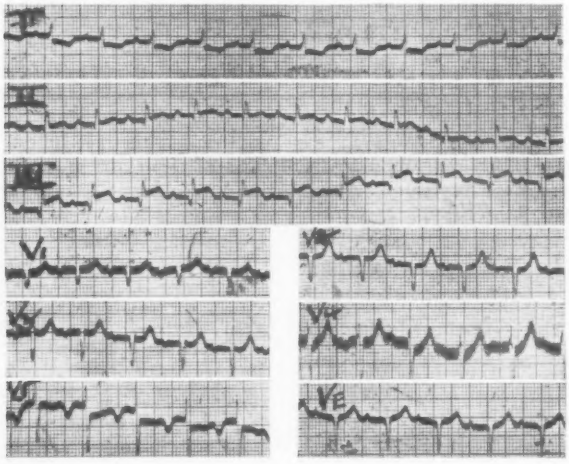

Fig. 2.—Case 2. ECG (July 18, 1947) showing Q_3 and elevated ST_3 consistent with posterior left ventricular infarction. Absent R waves in V_1, V_2, V_3.

Fig. 3.—Case 2. ECG (July 23, 1947) showing interference dissociation, Q_3 and inverted T_3, consistent with posterior left ventricular infarction.

Ocular fundi: tortuous arterioles and A–V nicking. Mouth: edentulous. Tongue dry. Chest: over the right lower lobe posteriorly there were impaired resonance, fine räles and suppressed breath sounds. Heart: point of maximal impulse in sixth left intercostal space 2 cm. from midclavicular line. Rate slow; rhythm regular. *No murmurs.* Abdomen: slight generalized tenderness and increased resistance to palpation. No masses. Liver thought to be percussed 4 cm. below right costal margin. Extremities: bilateral calf tenderness and 1+ pretibial edema, but Homan's sign negative. Two small purpuric spots were seen on the right leg. Neurologic: all deep reflexes were hyperactive. There was a questionable Babinski on the right. Pelvic and rectal: negative.

Laboratory Data. Hgb. 15 gm.; RBC 3,750,000; WBC 26,700 on admission; 94% polymorphonuclears and 6% lymphocytes. WBC 18,500 2 days before death. Cath. urine; sp. grav. 1025; Alb. 1+ no sugar; rare RBC, no WBC or casts. Stools guaic negative, yellow. Kahn, neg. BUN 60 mg. % on admission, later 70, 100, and 90 on day of death. CO_2 38 vol. % day of death. Blood culture negative. Prothrombin times 17 sec., later 24.5 and 24. 24.2 2 sec.

ECG. July 10, 1947. (Fig. 1.) Interference dissociation. Auricular rate 100; ventricular rate 60. QRS 0.06 sec., QT 0.32 sec.; Q3 present. July 12, interference dissociation. Q2 and Q3 present; ST2, ST3 elevated; ST1, ST4 depressed. July 16, same. July 18, 1947. (Fig. 2.) Sinus rhythm. PR 0.22 sec. QRS 0.08 sec. QT 0.36 sec. Q3 present. ST1 depressed, ST3 elevated. Q wave in V1, V2, V3, VE. ST in V5 depressed with T inverted. July 23, 1947. (Fig. 3.) Rate 100. PR 0.20 QT 0.26. Q3 present. ST3 elevated. ST1, 4 low, sagging. Nodal premature beat.

Chest Roentgen Ray. July 10, 1947. Heart 14.5 cm. in chest of 22.5 cm. Suggestive bilateral pulmonary edema. Area of increased density in right midlung field read as possible infarct.

Course. The patient was given penicillin for the first 10 days of her hospital stay. Her temperature became normal on the 3rd day and remained so except for elevations to 100° during the 3 days prior to death. On the morning following admission the heart rhythm was found to be irregular with pauses occurring every 2 to 8 beats. The runs of 2 to 8 beats seemed to be regular. On the 7th hospital day a harsh Grade 4 (Levine) systolic murmur was audible over the entire precordium. This murmur was loudest in the fourth left intercostal space just to the left of the sternum. (Fig.4). Phonocardiogram tracings taken on July 24

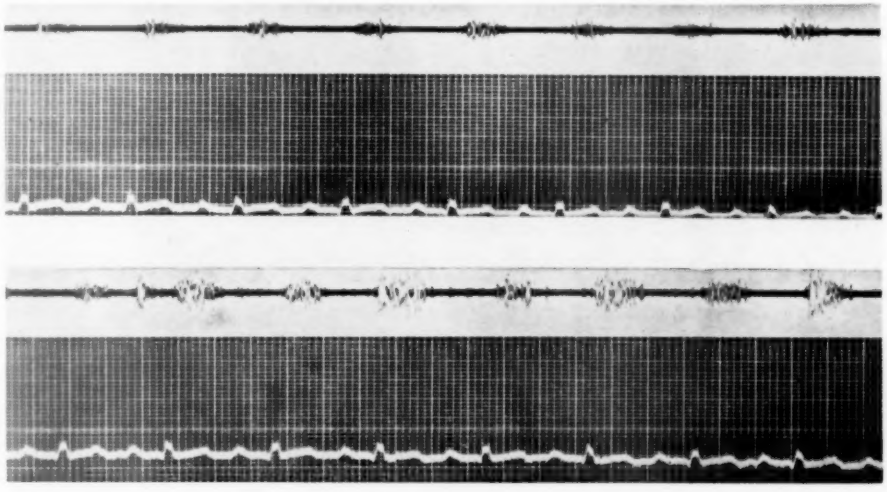

FIG. 4.—Case 2. Lower tracing taken at full volume. Upper tracing taken at approximately half volume

with microphone over fourth left intercostal space 3 cm. from sternum showed loud murmur filling nearly all of systole. A systolic thrill was felt in this area and at the cardiac apex. Two days prior to death the patient, who was previously only slightly improved, seemed worse. She became dyspneic with associated periods of apnea and began to run slight fever. On the 15th hospital day the patient was found to be unresponsive. B.P. 85/70. Respirations were 48. There was dullness and bronchial breathing over the right upper lobe. Lung bases were clear. Left border of cardiac dullness was 11 cm. from the midsternal line. The systolic thrill and murmur were present as before. There was no cyanosis. The patient expired later the same day.

Antemortem Diagnoses. (1) Posterior myocardial infarction; (2) probable perforation of infarcted interventricular septum; (3) probable bronchopneumonia or possible pulmonary infarction.

Necropsy Diagnoses. (1) Coronary atherosclerosis; (2) extensive progressive myocardial infarction; (3) old and recent mural thrombi; (4) *recent perforation of interventricular septum* through a fresh infarct; (5) multiple pulmonary emboli and pulmonary infarction; (6) chronic passive congestion of viscera with cardiac cirrhosis; (7) acute thrombosis of iliac, perivesical, uterine, right ovarian, renal veins; (8) squamous metaplasia of pancreatic duct with small foci of fat necrosis; (9) fibrosis of suprarenal capsule; (10) hemopericardium.

Gross Description of Heart (Figs. 5 and 6). The heart weighed 350 gm. The pericardial sac contained 24 cc. of grossly bloody fluid. The surface of the left ventricle showed petechial hemorrhages and large areas of pallor and yellowish discoloration. The epicardium was clouded in these areas and the underlying myocardium softened. Cardiac chambers were dilated with softening of the entire myocardium. The entire interventricular septum was soft, mushy, and yellow-gray as were the adjacent parts of both ventricles. There was also a section of the midportion of the left ventricular wall having the same consistency, all of these areas representing sites of old and

FIG. 5.—Anterior view of heart showing endocardial surface of left ventricle. Probe marks septal perforation. (Scale in inches.)

recent infarction. There was a large mural thrombus in the left ventricle and smaller mural thrombi in the right ventricle and right auricle. There was a complete perforation of the interventricular septum 1 cm. in diameter located 2 cm. below the mitral valve on the left side and just beneath the septal flap of the tricuspid valve on the right side. This perforation was 1.5 cm. from the epicardial surface of the left ventricle. It was ovoid in shape with irregular edges. The coronary arteries showed atherosclerosis.

FIG. 6.—Anterior view of heart showing endocardial surface of right ventricle. Probe marks septal perforation. (Scale in inches.)

matous and calcified plaques. The left coronary was occluded at its bifurcation by an atheromatous and calcified plaque. Similar luminal stenosis occurred 15 mm. distal to this in the left anterior descending branch. The right coronary ostium was narrowed by an atheromatous plaque in the aorta, but there was no area of occlusion apparent.

Discussion. In the 56 total cases, including the present, the time of survival after septal perforation was known in 38. Of these, 31 survived less than 1 month; these 31 had an average survival time of only 6.4 days. Six survived over 1 month but less than 1 year. One extraordinary case survived 4 years and 10 months.[17] Electrocardiographic findings were unknown in 38 cases; of the remaining 16, 4 showed posterior infarction; 4 "cardiac infarction," 1 anterior infarction, 1 complete block, 5 RAD, 1 RBBB, and 2 a large Q3. The signs were unknown in 10 cases; there were none in 2 cases. Of the 45 in whom physical findings were known, 43 showed systolic murmurs, usually maximal to the left of the lower sternum; 22 of the 45 showed systolic thrills; only 3 showed diastolic murmurs, and none showed diastolic thrills.

The 2 cases in which no murmur was heard were those of Bean,[2] and Bickel and Mozer.[4] The latter of these was a man 87 years old who had been well until 15 days before hospital admission. At this time he developed rather suddenly cough, dyspnea, anorexia and weakness.

He had no chest pain at any time. On admission he had Cheyne-Stokes breathing, heart sounds of poor quality, but a blood pressure of 150/70. He had signs of bronchopneumonia. No murmur was heard, and there is no mention of any pulmonary or cardiac condition which might obstruct the transmission of a murmur. The patient died on the 5th hospital day after an uneventful clinical course. At autopsy there were found thrombi obstructing both coronary arteries and a septal infarct with perforation. This perforation was in the posterior part of the septum and measured 2 x 0.5 cm.

Bickel and Mozer attributed the absence of any murmur in this patient to the large size of the septal perforation. In their discussion of the case they state that they regarded the cardiac tone and blood flow as adequate for the production of a murmur provided proper local conditions were present.

Bean's case also had a large septal perforation, but here the patient was in coma and shock and the blood pressure unobtainable. Among the other cases reviewed in the present paper were several with septal perforations as large or larger than Bean's, or Bickel and Mozer's cases, and in all of these murmurs alone or murmurs and thrills were found. In addition there appeared to be no correlation between the size or location of the perforation and the intensity of the murmur.

In Sager's[13] review, 15 of 18 cases gave a history typical of coronary occlusion. The physical findings, according to him, are those usually found in cases of patent interventricular septum. When both of these conditions occur in the same patient, the diagnosis of perforation of the infarcted septum is to be made. The murmur is usually loud, according to Sager, usually accompanied by a thrill, and transmitted to the left axilla but not to the neck vessels. He explains the infrequency of septal perforation as compared to that of rupture of the ventricular wall by the rich arterial supply to the septum.

According to Weber,[15] the diagnosis of perforation of the infarcted septum is made by the sudden appearance of a loud systolic murmur and a palpable thrill in a patient who is known to have myocardial infarction. He states that the prognosis is much worse in this condition than in the usual case of myocardial infarction, not because the perforation interferes with cardiac dynamics, but because the perforation is ordinarily seen only in cases of massive infarction. He states that the left coronary artery is usually occluded in these cases, and that in addition the right coronary is usually sclerosed and incapable of aiding in anastomoses. Lober and Hertzog[9] state that symptoms at the time of perforation are unusual, but if the patient lives long enough, there always develops an intractible right-sided heart failure, death from heart failure being inevitable. Wood and Livezey[17] make the same statement.

The conditions to be differentiated from perforated septum are rupture of a papillary muscle of the left ventricle following cardiac infarction, congenital potency of the interventricular septum, perforation of the septum due to subacute bacterial endocarditis, and rupture of a mitral chorda tendinea.

With rupture of a papillary muscle complicating infarction, the heart, according to Sager,[13] usually becomes markedly dilated, the murmurs are even louder, and diastolic murmurs are more common. Too, the signs are evenly distributed over the precordium, or maximal at the apex. Weber states that here there is a more sudden change for the worse, and more likelihood of left-sided than right-sided heart failure. Until July 1946, only 16 cases[6,7,8,10,12,14] of ruptured papillary muscle following cardiac infarction are recorded with physical examination of the heart. Of these, 6 had an apical systolic murmur, 1 had an apical diastolic murmur, 3 had an apical systolic and diastolic murmur; 1 had "murmurs," 1 had "noises," 1 had a friction rub in the

fourth left interspace and an apical gallop; in 2 no murmurs were heard, and in 1 case there was no mention of murmurs. The prognosis in this condition is approximately that of the former one. Of 23 reported cases of ruptured papillary muscle which can be attributed to coronary occlusion, only 1 survived longer than 1 month. In the remaining cases the average survival was 4 days.

Congenital patency of the interventricular septum may cause confusion, but should be easily differentiated by the absence of history or ECG changes of myocardial infarction. It would presumably be impossible properly to diagnose a patient having a previously undiagnosed congenitally patent septum complicated by myocardial infarction. Acquired perforation of the septum due to subacute bacterial endocarditis is to be differentiated by the positive blood cultures and the presence of embolic phenomena. Perforation of the infarcted septum must also be differentiated from ruptured chordæ tendineæ, according to Bailey and Hickam.[1] The latter condition is to be suspected in a middle-aged or elderly individual who suddenly, without dramatic incident, develops a loud systolic murmur, usually with a thrill, maximal at the cardiac apex and along the left sternal border. Congestive heart failure does not usually appear in these cases for months or years. The absence of clinical or laboratory evidence of cardiac infarction serves to differentiate ruptured chordæ from acquired patency of the infarcted interventricular septum.

Summary. Two cases of perforation of the infarcted interventricular septum are reported in 1 of whom the diagnosis was made antemortem.

A total of 56 such cases have been found in the literature; 15 of these were diagnosed antemortem. In 38 whose survival time was known, 31 survived less than 1 month, 37 less than 1 year, and 1 survived 4 years and 10 months. Forty-three cases of 45 examined showed systolic murmurs, usually maximal to the left of the lower sternum; 22 showed systolic thrills; only 3 showed diastolic murmurs.

The diagnosis should be suspected in any patient known to have a myocardial infarction who suddenly develops a systolic murmur and thrill to the left of the lower sternum. The only condition likely to be confused is rupture of a papillary muscle following cardiac infarction, where the condition of the patient suddenly becomes worse, the murmurs are louder and nearer the apex, and the ensuing failure is left-sided rather than right-sided.

Authors' Note: Since the submission of this article for publication 2 additional cases of rupture of the interventricular septum as a complication of myocardial infarction have been reported. One case was reported by D. Carroll and S. D. Cummins: Am. Heart J., **34**, 893, 1947. The other by R. S. Diaz-Rivera and A. J. Miller: Am. Heart J., **35**, 126, 1948. The first case showed a loud apical systolic murmur, heard also over precordium and the left upper quadrant. The second showed an apical systolic murmur heard also in the third left intercostal space. No diastolic murmur or systolic thrill was described in either case.

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THE USE OF VITAMIN E IN HEART DISEASE

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Early in 1946 attention was called by Vogelsang and Shute[11] to the marked improvement resulting from the administration of vitamin E to patients with various forms of heart disease. Considerable publicity was given to the phenomenal therapeutic results claimed in congestive heart disease and angina pectoris by those authors. Evan Shute[10] was quoted as saying: "We have not learned of a single failure. The percentage of success is remarkable." Patients began to request the treatment from physicians, and less scrupulous drug houses quoted entire paragraphs from the articles recommending vitamin E, in their attempts to sell preparations of alpha-tocopherol. Editorial comment in the *Journal of the American Medical Association*,[2] however, decried this publicity given to unconfirmed work, stating "nothing in the known pharmacological actions of vitamin E would lead one to expect either a vasodilating action, a myotonic effect or an ability to repair damaged heart muscle in human beings."

Because of the widely divergent views quoted above, we decided to investigate the influence of vitamin E on various cardiac conditions. Preliminary studies were begun in August, 1946, and we are reporting here our observations on the first 25 patients studied.

Plan of Study. Cases for this study were selected from the Cardiac Clinic of the Jewish Hospital and the private practice of one of us (S. B.). After a complete history, physical examination, blood count, blood Wassermann, urinalysis, electrocardiogram and orthodiagram, a brief period of clinical observation was carried out. The patients were then given orally 400 mg. of vitamin E* daily in divided doses. Within 2 to 4 weeks this dose was reduced to 300 mg. daily. The patients were then followed for 3 to 6 months, whenever possible. Electrocardiographs were repeated at 2- to 4-week intervals. Those patients previously receiving such preparations as digitalis, mercurials, phenobarbital, and the like, were continued on their medication so that we could more accurately evaluate the effect of the vitamin E. In some cases after the vitamin E was given, either digitalis or mercurials were then discontinued, to determine if recurrence of the cardiac failure would be prevented. In some of those patients exhibiting apparent symptomatic response to vitamin E therapy, similarly appearing tablets of calcium gluconate were substituted as a placebo.

Results. Of the 25 patients treated with vitamin E, 22 were considered to have been adequately followed, and form the basis of this report. Of these, 11 had congestive heart failure and 5 angina pectoris. The remaining 6 had hypertension and/or arteriosclerotic heart disease, without failure or angina. The essential features of these 3 groups of cases are shown in Tables 1, 2 and 3 respectively. A summary of the therapeutic results obtained is presented in Table 4.

Congestive Failure (Table 1). Of the 11 patients with congestive heart failure, 3 are listed as questionably improved. In Case No. 2, there was diminution in the frequency of attacks of paroxysmal nocturnal dyspnea. However the patient had discontinued working in a

* Vitamin E in the form of 100-mg. tablets of Ephynal Acetate was kindly provided for this study by Hoffman-La Roche Company.

Table 1.—Data in 11 Patients With Congestive Failure Treated With Vitamin E

Failure

TABLE 1.—DATA IN 11 PATIENTS WITH CONGESTIVE FAILURE TREATED WITH VITAMIN E

Case	Age	Sex	Failure: Severity	Failure: Clinical features	Clinical diagnosis	Electrocardiogram	Orthodiagram	Daily dose of vitamin E	Results
1	32	F	Mild	Exertional dyspnea	Rheum. h.d.; aort. st. and reg.; mitr. st. and reg.	Auric. extrasystoles; auric. dis.	Normal size; contour of mitral stenosis	400 mg. 3 wks. 300 mg. 22 wks.	After vitamin E given for 2 mos. dyspnea increased, necessitating digitalization; subsequent relief
2	60	F	Mod.	Paroxysmal dyspnea; on digitalis	Hypert. h.d.	Myocard dis.; dig. effects	Normal size; shape suggests left ventricular hypertrophy; tortuous aorta	400 mg. 5 wks. 300 mg. 11 wks.	Moderate symptomatic improvement but patient stopped working in factory during course of study
3	58	F	Mod.	Dyspnea, edema; on digitalis	Rheum. h.d.; mitr. st. and reg.	Auric. fib.; myocard. dis.; dig. effects	30% enlargement; dilated pulm. art.; enlarged l auric.	400 mg. 6 wks. 300 mg. 19 wks.	Increase of failure after being on vitamin E for 6 mos.; mercurial diuretics necessary
7	65	F	Mild	Exertional and nocturnal dyspnea	Hypert. h.d.	Normal	20% enlargement; tortuous aorta	400 mg. 9 wks. 300 mg. 14 wks.	Decrease in dyspnea when on vitamin E; effect duplicated by placebo
10	67	M	Marked	Dyspnea and precordial discomfort; on digitalis	Hypert. and a–s. h.d.	Auric. fib.; ventric. extrasyst.; intravent. cond. defect; l. vent. hypert.	70% enlargement	400 mg. 19 wks. 300 mg. 4 wks.	Marked subjective improvement shortly after starting vitamin E; rapid progress of failure when digitalis was stopped, while vitamin E was continued; re-control of failure by digitalis; further subjective improvement with placebo
11	44	M	Mild	Asymptom.; on digitalis since failure in 1936	Rheum. h.d.; mitr. st. and reg.	Auric. fib.; dig. effects	40% enlargement; dil. pulm. art.; dil. l. auric.	400 mg. 8 wks. 300 mg. 5 wks.	No effect noted; stopped attending clinic after 13 wks.
15	71	M	Marked	Dyspnea; edema; hepatomeg.; on digitalis and mercurials	Hypert. and a–s h.d.	Auric. fib.; complete heart block; myocard. dis.; dig. effects	74% enlargement	400 mg. 2 wks. 300 mg. 4 wks.	Congestive failure progressed after taking vitamin E for 6 wks. (along with digitalis and mercurials); hospitalization necessary; response to bed rest, digitalis, diuretics
18	47	F	Marked	Dyspnea, anasarca; on digitalis and mercurials	Rheum. h.d.; mitr. st. and reg.; tricusp. reg.	Large P waves; dig. effects	Marked enlargement*	400 mg. 3 wks.	Mercurials stopped after vitamin E given for 2 wks.; failure became more marked
20	62	F	Marked	Dyspnea, edema; on digitalis and mercurials	Hypert. and a–s. h.d.	Left bundle branch block; myocard. dis.	68% enlargement	400 mg. 5 wks. 300 mg. 8 wks.	Felt better while taking vitamin E and mercurials were discontinued; however, at this time she was in bed because of acute thrombophlebitis; on resuming activity failure progressed, necessitating further use of mercurials, despite continuation of vitamin E
21	18	M	Marked	Dyspnea, anasarca; on digitalis and mercurials	Rheum. h.d.; mitr. st. and reg.; tricusp. st.	Rt. axis dev.; auric. fib.; myocard. dis.; dig. effects	Marked enlargement*	400 mg. 2 wks. 300 mg. 3 wks.	Mercurials stopped when vitamin E begun; failure gradually became more marked
22	43	F	Marked	Dyspnea, anasarca; on digitalis and mercurials	Hypert. h.d.	Normal	Mod. enlargement*	400 mg. 10 days	No improvement with vitamin E in addition to digitalis and mercurials; later response to digitalis and mercurials alone

* Teleoroentgenogram; patients unable to stand for orthodiagram.

TABLE 2.—DATA IN 5 PATIENTS WITH ANGINA PECTORIS TREATED WITH VITAMIN E

Case	Age	Sex	Severity of angina	Clinical diagnosis	Electrocardiogram	Orthodiagram	Daily dose of vitamin E	Results
9	77	F	Mod.	Hypert. h.d.	L. ventric. hypert., old post. infarction	30% enlargement	400 mg. 3 wks. 300 mg. 4 wks.	No improvement
13	62	F	Mod.	Hypert. h.d.	Previously: left ventric. hypert.; intraventric. cond. defect; prob. antero-septal damage. Just prior to study: left bundle branch block	45% enlargement	400 mg. 7 wks. 300 mg. 17 wks.	No improvement; left bundle branch block, present just before vitamin E started, disappeared for several months but reappeared transitorily while vitamin E was still being taken
14	69	M	Mod.	A–s. h.d.	Normal	Normal	400 mg. 2 wks. 300 mg. 4 wks.	Questionable slight decrease in frequency of attack
17	75	M	Mod.	Hypert. h.d.	L. ventric. hypert.	57% enlargement	400 mg. 3 wks. 300 mg. 12 wks.	Slight improvement in exercise tolerance, which occurred however coincidentally with milder weather; no change when vitamin E discontinued
19	41	M	Mod.	Myocard. dis.	Normal	Normal	400 mg. 3 wks.	Three weeks after beginning vitamin E angina increased and myocardial infarction developed; confirmed by electrocardiographic changes

TABLE 3.—DATA IN 6 PATIENTS WITH HYPERTENSION AND/OR ARTERIOSCLEROTIC HEART DISEASE TREATED WITH VITAMIN E

Case	Age	Sex	Clinical features	Clinical diagnosis	Electrocardiogram	Orthodiagram	Daily dose	Result
4	28	M	Nervousness; b.p. 190/120	Essential hypertension	Normal	Normal	400 mg. 5 wks. 300 mg. 25 wks.	No effect noted
5	73	M	Myocard. infarction in 1944; slight dizziness on effort	Hypert. and a–s h.d.	Myocard. dis. intraventric. cond. defect; healed ant. and post. infarctions	70% enlargement	400 mg. 4 wks. 200 mg. 11 wks.	No effect noted
6	47	M	Myocard. infarction in Feb. 1946; vague anterior chest discomfort	A–s. h.d.	Healed antero-lateral infarction	20% enlargement; aorta increased in density	400 mg. 4 wks. 300 mg. 22 wks.	No effect noted
8	58	F	Dizzy spells; hot flushes; b.p. 220/120	Hypert. h.d.	L. ventric. hypert.; myocard. dis.	30% enlargement	400 mg. 1 wk. 200 mg. 2 wks. 300 mg. 16 wks.	No effect noted
12	55	F	Headaches; b.p. 240/130	Hypert. h.d.	L. ventric. hypert.; myocard. dis.	25% enlargement	400 mg. 4 wks. 300 mg. 17 wks.	Subjective improvement (headaches); effect duplicated by placebo; b.p. unchanged.
16	72	M	Dyspnea on effort; b.p. 210/100	A–s. and hypert. h.d.	Probably some myocard. dis.	40% enlargement	400 mg. 4 wks. 300 mg. 2 wks.	No effect noted

factory at about the time of her symptomatic improvement, so that it was difficult to determine whether the medication or the decreased activity was responsible. Case No. 7 displayed a decrease in dyspnea, but this effect was also produced by the administration of a placebo. Case No. 10 claimed marked lessening of his dyspnea and precordial discomfort within a week after beginning vitamin E. However, when digitalis was stopped while vitamin E was continued, signs of cardiac failure appeared (increased dyspnea, tachycardia, pulmonary congestion). Compensation was restored with digitalis, and when a placebo was administered, the patient again claimed marked improvement.

Table 4.—Results of Treatment of 22 Patients With Heart Disease With Vitamin E

	Cardiac	Angina	Hypertension and/or arteriosclerotic heart disease	Totals
Markedly improved	0	0	0	0
Definitely improved	0	0	0	0
Questionably improved	3	2	1	6
Unimproved	5	2	5	12
Worse	3	1	0	4
	11	5	6	22

The remaining 8 cases were either unimproved or became distinctly worse during therapy with vitamin E. In 3 cases (Nos. 18, 20, 21), mercurial diuretics, which previously had been administered regularly, were discontinued during our treatment with vitamin E, in order better to evaluate the effectiveness of the therapy. In all these instances the cardiac failure progressed and the mercurials again had to be given. Three of these 8 patients became distinctly worse while taking vitamin E. Patient No. 1 developed increasing dyspnea and did not obtain relief until digitalis therapy was instituted. In Case No. 3, edema and dyspnea increased after the patient had been on continuous vitamon E therapy for 6 months; mercurial diuretics were then effectively employed. Patient No. 15, who had previously suffered several bouts of severe congestive failure, developed such an episode after he had been taking vitamin E for 6 weeks. Hospitalization and complete bed rest became necessary.

In none of the 11 patients with congestive heart failure was a definite therapeutic effect of the vitamin E noted.

Angina Pectoris (Table 2). Of the 5 patients with angina pectoris, 2 are listed as questionably improved. In Case No. 14 there was possibly a slight decrease in the frequency of anginal seizures. Patient No. 17 claimed slight improvement in exercise tolerance. This occurred, however, coincidentally with the arrival of milder weather. There was no increase in symptoms when the vitamin E was discontinued. Two patients (Nos. 9 and 14) showed no improvement and 1 (No. 19) became distinctly worse. He developed an acute myocardial infarction 3 weeks after the vitamin E was begun.

In none of these 5 patients with angina pectoris was there unequivocal evidence of improvement during treatment with vitamin E.

Hypertension and/or Arteriosclerotic Heart Disease (Table 3). Of the 6 patients in this group, none exhibited a definite therapeutic response to vitamin E. In 1 (No. 12) there was a decrease in the incidence and severity of the headaches, but this effect was duplicated by a placebo.

Effect on Blood Pressure, Electrocardiogram and Orthodiagram. There was no definite alteration in the blood pressure of any of the patients. Serial electrocardiograms revealed no significant changes in any case. Case No. 13 illus-

trates the transitory nature of certain changes unrelated to therapy. A left bundle branch block present just prior to the beginning of this study, disappeared for several months during the administration of vitamin E. It reappeared, however, and again disappeared while the drug was still being taken. There were no changes in the orthodiagrams, except for the slight increase in heart size of several patients while their failure was becoming more severe.

Discussion. A study such as this is open to a number of criticisms. The series of cases presented is small. Furthermore, in evaluating the effect of a drug in congestive heart failure or angina pectoris, it would have been preferable to present data obtained more objectively, such as circulation times, exercise tolerance tests, venous pressure, and vital capacity. We had indeed intended expanding our studies to include these observations, but the discouraging results presented in this preliminary report deterred us from carrying these investigations further.

A consideration of the literature reveals little basis for recommending the use of vitamin E in human heart disease. The early experimental observations included the effects of vitamin E deficient diets on a cow,[5] rabbits,[6] chicks[1] and rats,[7] and on isolated heart muscle strips.[4] These results have not been uniform. For example, one observer[7] reported cardiac disturbances in rats fed vitamin E deficient diets. Another,[3] however, found no striking change in the cardiograms of rats fed vitamin E deficient diets for 1 year.

Even if we accept the debatable conclusion that vitamin E deficient diets in animals produce cardiac abnormalities, it does not appear logical to assume that vitamin E would therefore be of value in the treatment of such varied cardiac disturbances in human beings as rheumatic heart disease, angina pectoris and congestive failure. At no time has there been any demonstrable proof that patients with cardiac failure or angina pectoris are deficient in vitamin E. It would appear just as illogical to recommend thiamin chloride or calcium salts or thyroid for all patients with heart disease, because prolonged deprivation of these agents may be associated with clinical or electrocardiographic changes.

The only publications to date attesting to the beneficial effects of vitamin E in humans have been those of Vogelsang, Shute and Shute.[8,9,12,13,14] Certainly if one reviews their articles critically, one hesitates to recommend vitamin E with the degree of enthusiasm they have manifested. As a matter of fact, some of the data they present hardly warrant the conclusions reached. In one of their papers,[12] for example, they present summaries of 10 cases treated with vitamin E. In Case No. 2, they state: "she insisted on stopping the dose of ephynal on March 29, and reverted to her digitalis instead. Her relief was immediate." In Case No. 3, the regression of electrocardiographic changes in a patient with acute rheumatic fever are attributed to vitamin E therapy, rather than the natural course of the disease. In Case No. 6, the description of the cardiograms suggests acute myocardial infarction. Decrease of the chest pain need not have been due to vitamin E. Many patients with acute myocardial infarction have less pain on the 2nd day than the first. In Case No. 8, the improvement was attributed to vitamin E after a month of vitamin E therapy, even though digitalis was begun 2 weeks before improvement was noted.

In their most recent paper Vogelsang, Shute and Shute[14] present a summary of their findings in 126 unselected cases of heart disease, and discuss some of the underlying pathologic physiology. They stated that improvement following therapy with vitamin E occurred because vitamin E (*a*) affects the blood-vessels, (*b*) affects the myocardium, (*c*) may influence the pacemaker of the heart, and (*d*) may influence the thrombotic features of coronary artery disease by improving the col-

lateral circulation in the ischemic area. We are not familiar with any proof of these mechanisms in the experimental animal or in man. They concluded that the great majority of cases with angina, rheumatic heart disease, congestive failure and hypertension are helped considerably by the administration of vitamin E.

Our results are so at variance with those published that we hesitate to recommend the use of vitamin E in cardiac disease. Certainly much more scientific work must be done in this field before suggestions are made that so appreciably alter our present forms of cardiac therapy.

Conclusions. 1. Vitamin E in 300 to 400 mg. doses daily was used in the treatment of 11 patients with congestive heart failure, 5 patients with angina pectoris and 6 patients with hypertension and/or arteriosclerotic heart disease.

2. In no case was there any demonstrable effect on the electrocardiogram, orthodiagram or blood pressure.

3. None of the 22 patients was markedly or moderately improved. Six patients were questionably improved and the remainder showed no change or became distinctly worse.

4. Our results were not encouraging enough to recommend the use of vitamin E in the treatment of angina pectoris, congestive failure or hypertension.

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OSTEOPOROSIS OCCURRING DURING POTASSIUM THIOCYANATE THERAPY FOR HYPERTENSIVE DISEASE

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After the extensive use of potassium thiocyanate in the treatment of hypertension, occasional instances of osteoporosis and arthralgia were noted at this clinic. Since no previous description of this phenomenon could be found, a further investigation was undertaken.

The records of 5000 consecutive patients with hypertension who had been seen on two or more occasions at the Mayo Clinic in the period 1939 to 1944, inclusive, were reviewed. Potassium thiocyanate had been given to 360 patients of this group. Unexplained osteoporosis occurred in 7. Since 1944 there have been 4 additional patients with this syndrome. No history of trauma or injury at onset could be elicited from these patients. One or more extremities, usually the lower, were involved. Patients with involvement of the spine or pelvis were not included, as senile osteoporosis could not be excluded. Similarly omitted were those who had a diagnosis of chronic infectious arthritis, periarthritis, rheumatoid arthritis or fibrositis. Unexplained osteoporosis was not observed in the group of more than 5000 patients with hypertension who were not receiving potassium thiocyanate.

Clinical Data. A summary of pertinent clinical data is given in Table 1. The incidence of osteoporosis in the group of patients who were receiving potassium thiocyanate therapy was approximately 2%. There were 6 women and 5 men. The age ranged from 46 to 68 years (average 56.5). The dosage of the drug varied considerably during the course of treatment, but was usually in the range of 6 to 9 grains (0.4 to 0.6 gm.) daily. Onset of symptoms associated with the osteoporosis generally occurred in 3 to 6 months after administration of the drug was started. They consisted of (1) pain on use of the extremity which began insidiously and gradually increased in severity, and (2) subsequent mild swelling of the joint or joints involved, but with no acute inflammatory reaction. The severe cases simulated those of extensive posttraumatic osteoporosis. Roentgenograms, which were limited to the involved regions, revealed mild to marked diffuse osteoporosis.

Active therapeutic measures directed toward the osteoporosis were carried out in 7 cases while thiocyanate therapy was being continued. These consisted of physiotherapy, active and passive movement, shoe corrections, walking-casts, elastic bandages, and preparations of calcium and phosphate for oral use. Symptoms continued to progress despite these measures in 6 cases. Slight improvement

* Abridgment of thesis submitted by Dr. Hinchey to the faculty of the Graduate Hospital of the University of Minnesota in partial fulfillment of the requirements for the degree of Master of Science in Orthopedic Surgery.

TABLE 1.—OSTEOPOROSIS OCCURRING DURING POTASSIUM THIOCYANATE THERAPY FOR HYPERTENSIVE DISEASE: CLINICAL DATA

Case	Age (years)	Sex	Potassium thiocyanate grains daily (average)	Blood pressure (mm. of mercury)		Onset of symptoms (months*)	Part of body involved	Condition after specific therapy	Time for improvement (months)
				Before therapy	During therapy				
1	49	F	6	159/120	150/110	6	Right lower extremity	Worse after 7 months	6
2	46	M	6†	228/174	170/105	4	Right hip	..	10
3	47	M	9	219/130	210/130	4	Both ankles, right hip and knee	Worse after 5 months	3‡
4	53	M	9§	225/125	220/120	4	Feet and ankles	Worse after 5 months	Unknown, complete recovery
5	55	F	4	270/130	236/120	20	Forearms, wrists, left knee	..	Unknown, complete recovery
6	66	F	6¶	220/110	206/110	3	Ankles	..	7
7	50	F	6	218/134	190/110	4	Feet and ankles	Improved after 18 months	12
8	62	M	6	160/90	164/90	3	Left lower extremity	Worse after 2 months	4
9	68	F	6	190/100	184/100	5	Feet and ankles	Worse after 4 months	5
10	60	M	6	..	190/108	6	Right foot and ankles	..	5
11	66	F	6	..	188/98	5	Left lower extremity	Worse after 7 months	3‡

* Interval between beginning of administration of potassium thiocyanate and onset of symptoms.
† Five days a week.
‡ Improving, but not completely asymptomatic.
§ Six days a week.
¶ Four days a week.

over a period of several months was noted in the 7th case under such measures, and the rate of improvement was accelerated when the drug was stopped.

Cessation of potassium thiocyanate therapy was followed by relief in every one of the 11 cases whether or not specific measures of treatment were used. Improvement was generally evident in 2 to 3 months; recovery was complete in 5 to 7 months. Administration of the drug was resumed in 4 instances. In 2 cases there was no recurrence of symptoms or of osteoporosis. Symptoms recurred in the other 2 but were again relieved when thiocyanate therapy was once more discontinued.

Laboratory Data. The laboratory findings are listed in Table 2. The blood thiocyanate levels shown were those present at the time of admission with the osteoporosis. The thiocyanate levels in general varied considerably in different patients and in the same patient from time to time.

TABLE 2.—OSTEOPOROSIS OCCURRING DURING POTASSIUM THIOCYANATE THERAPY FOR HYPERTENSIVE DISEASE: BLOOD CHEMICAL FINDINGS

Case	Mg. per 100 cc.					Alkaline phosphatase, Bodansky units
	Cyanates	Urea	Serum calcium	Serum phosphate	Uric acid	
1	4.7	34				
2	9.3	24	10.0	2.9	3.5	3.7
3	3.8	22				
4	8.9	28	9.8	..	4.4	
5	7.6	24				
6	11.9	28				
7	8.5	28	..	..	3.3	
8	11.6	30	8.7	2.2	..	2.8
9	9.8	30	8.7	3.1		
10	..*	30				
11	4.4†	38	9.4	2.8	1.8	3.1

* Drug had recently been discontinued; no level ascertained.
† Level 2 weeks after drug had been discontinued.

The value for blood urea was within normal limits in all cases. The blood uric acid level was normal in 4 cases. The sedimentation rate was elevated in 2 of the 11 cases, but remained so after complete recovery in both. The serum phosphate level was determined in 4 patients and the blood alkaline phosphatase in 3. All were within the usual range of normal. The serum calcium level, determined in 5 instances, was within normal limits in 3 and slightly below normal in 2. In one of these, Case 9, a urinary Sulkowitch test gave a Grade 4 reaction (graded on the basis of 1 to 4, in which 1 represents the least, and 4 the greatest concentration of calcium).

Report of Cases. The numbers of the following 3 cases, which are reported in detail, correspond to the case numbers in Tables 1 and 2.

CASE 2. A white man, 46 years of age, registered at the clinic in January, 1939, for consultation regarding hypertension. His blood pressure was 228 mm. of mercury systolic and 174 diastolic. Potassium thiocyanate therapy was instituted; the dosage was 6 gr. (0.4 gm.) a day, 5 days a week.

He returned July 18, 1939, complaining of localized pain in the right hip. Its onset in May had been quite sudden, and the pain had been continuous and progressive since then. Only the use of crutches had provided some relief.

The patient appeared in moderate distress and walked with a noticeable limp. Atrophy of the right buttock and quadriceps was noted. His blood pressure was 170 mm. of mercury, systolic, and 105, diastolic. Roentgenograms of the pelvis and right femur revealed marked osteoporosis of the right acetabulum and upper part of the femur. The blood chemical findings are given in Table 2.

Potassium thiocyanate therapy was discontinued; treatment consisted of the oral

administration of calcium and viosterol and the patient continued to use crutches. In October, 1939, he was much improved. A roentgenogram at that time revealed some residual osteoporosis.

Recovery was complete in May, 1940, and roentgenograms revealed no osteoporosis. Potassium thiocyanate therapy was resumed in November, 1940, and continued until the patient died more than 5 years later. There was no return of symptoms.

CASE 3. A white man, 47 years of age, was admitted to the clinic in October, 1942, complaining of severe headaches. His blood pressure was 219 mm. of mercury, systolic, and 130, diastolic. Nine grains (0.6 gm.) daily of potassium thiocyanate was prescribed, beginning October 20, 1942.

In July, 1943, he was readmitted. His headaches had improved. However, he complained of pain and swelling in the right knee which had begun early in 1943 (Fig. 1) and which had been followed in a month by similar symptoms in the right ankle. Heat, massage and sun-lamp therapy had failed to provide any relief. He had discontinued use of the thiocyanates in April and had had the ankle strapped in May; shortly thereafter he noted improvement. He had then resumed thiocyanate therapy and his symptoms returned. At that time the symptoms were located in the right knee and hip.

General physical examination gave essentially negative results except for the hypertension and a limp favoring the right leg. His blood pressure was 210 mm. of mercury, systolic, and 130, diastolic. Roentgenograms of the right hip and knee revealed moderate diffuse osteoporosis (Fig. 2). The blood cyanate level was 3.8 mg. per 100 cc.; the blood urea was 22 mg. per 100 cc.

Baking, massage and exercises for hip, knee and ankle were prescribed. The dosage of potassium thiocyanate was increased to 12 to 15 gr. (0.78 to 1 gm.) daily. The patient wrote in September, 1943, that he was worse and that both ankles were now involved. Potassium thiocyanate therapy was once more discontinued. A letter from the patient in December, 1943, stated that he

FIG. 1.—(Case 3.) Appearance of right knee on March 8, 1943, 5 months after potassium thiocyanate therapy had been started.

was much better and was steadily improving.

CASE 9. A white woman, 68 years old, was seen at the clinic in January, 1945, because of severe headaches. Her blood pressure was 190 mm. of mercury, systolic, and 100, diastolic. Potassium thiocyanate therapy was begun at that time. The dosage varied, but averaged 6 to 9 gr. daily.

The roentgenograms revealed diffuse osteoporosis of both feet and ankles (Fig. 3*a*). The blood chemical findings are given in Table 2. The reaction to the urinary Sulkowitch test on a 24 hour specimen was graded 4.

The patient was instructed to discontinue all therapy, including the use of thiocyanates

FIG. 2.—(Case 3.) Appearance of right knee July 26, 1943. Increased osteoporosis despite intensive local therapy is apparent while the patient was still on potassium thiocyanate.

The patient returned in October, 1945, stating that her headaches were relieved but that her feet and ankles were painful, swollen and stiff. Pain had begun in June, 1945, followed shortly by the stiffness and swelling. Her symptoms had since become progressively worse despite physiotherapy and the use of contrast baths, shoe corrections and elastic bandages.

General physical examination gave essentially negative results except as regards the hypertension and the condition in lower extremities. The blood pressure was 184 mm. of mercury, systolic, and 100, diastolic. Slight diffuse swelling, stiffness and tenderness of both feet and ankles were noted.

as well as measures directed at the osteoporosis. She stated, on her return in April, 1946, that her symptoms had steadily improved and that she was bothered only occasionally. Disappearance of the osteoporosis was evident on roentgenologic examination (Fig. 3*b*).

Comment. No cause for this syndrome other than thiocyanate therapy could be found. Trauma at the onset was consistently denied. One patient sprained the involved ankle shortly after the initial appearance of symptoms and this sprain markedly aggravated her discomfort. No other therapeutic procedure was instituted

for any significant period of time during the development of the osteoporosis in any case. Concurrent, but probably unrelated, conditions occurred in 2 patients. One consisted of a diarrhea beginning 2 months after the onset of symptoms. The other was a burn of the thumb with secondary infection. Sulfathiazole treatment was administered to the latter patient in the interval just preceding the appearance of symptoms; however, the osteoporosis in this case involved the ankles. No evidence of renal insufficiency was noted in any of these patients. The possibility of a local vascular accident with overcompensating collateral circulation and hyperemia was suggested as a cause for the osteoporosis. The absence of this syndrome in more than 5000 consecutive patients with hypertension who did not receive potassium thiocyanate makes it extremely unlikely that a complication of the hypertensive disease was the underlying cause of the osteoporosis.

FIG. 3.—(Case 9.) *a*, Osteoporosis of right foot and ankle which occurred while the patient was receiving potassium thiocyanate; the condition continued despite intensive local measures directed at the osteoporosis itself. *b*, Improvement after discontinuance of potassium thiocyanate therapy and of that for osteoporosis.

The following evidence suggests that the osteoporosis was a result of thiocyanate therapy: (1) there was a sufficient time interval between institution of the drug and onset of symptoms; (2) improvement was noted in a reasonable period of time after discontinuance of the drug, whether or not therapy was directed at the osteoporosis; (3) in 6 of the 7 cases the usual specific therapeutic measures failed to halt progression of the osteoporosis so long as potassium thiocyanate therapy was continued; (4) increase in dosage of the drug in one instance resulted in an aggravation of symptoms within a few weeks; and (5) resumption of use of the drug after recovery produced recurrence of symptoms in 2 of 4 cases, with relief after the drug was again discontinued.

A mode of action of the potassium thiocyanate must still be postulated. No marked lowering of blood pressure was noted consistently. The osteoporosis occurred far too infrequently to be the result of a direct action of the drug on bone itself.

Potassium thiocyanate therapy has been used in more than 100 cases of severe migraine without the production of any syndrome such as that of osteoporosis. Evidence at hand at present seems to favor a slight but prolonged interference with calcium metabolism as the mechanism of the production of the osteoporosis. The actual dosage of the drug is small, although it is in excess of those of aluminum and iron which have been recommended in different types of bone malacia. In the presence of adequate calcium intake and normal metabolism, such a slight interference with calcium metabolism would scarcely be sufficient to produce osteoporosis, and it has not occurred in the migrainous patients who represent a younger age group. However, should there be a tendency to a negative calcium balance, which is so often found in individuals of the age group with which this report deals, then any additional deficiency in calcium metabolism could produce such a picture. The finding of subnormal values for serum calcium in 2 cases and of a Grade 4 reaction to the Sulkowitch test in 1 case was further suggestive evidence of this mechanism. Thus all known factors, including the time element both for production of and recovery from this syndrome, the infrequency of occurrence of the syndrome, the dosage of the drug, and the group of individuals dealt with, were consistent with a minor additional interference with calcium metabolism. Further investigation along these lines is being carried out at present.

Regardless of the mechanism of production of osteoporosis, one should be cognizant of the occasional occurrence of this condition and should insure adequate calcium intake in individuals who are taking potassium thiocyanate. The use of this drug in the presence of a fracture may be inadvisable, not as regards union of the fracture, which is a local affair, but because any interference with calcium metabolism in the presence of an increased calcium requirement might precipitate osteoporosis. Potassium thiocyanate therapy may similarly be contraindicated in the presence of bone malacia, such as senile osteoporosis or osteitis deformans, as use of the drug could cause further increase of decalcification and aggravation of symptoms.

Summary and Conclusions. Osteoporosis with arthralgia was noted in 2% of patients receiving potassium thiocyanate therapy for hypertension. No similar syndrome was found in more than 5000 consecutive cases of hypertension in which this drug was not used. Evidence at hand seems to suggest slight and prolonged interference with calcium metabolism as a possible mechanism for production of the syndrome. It appears that adequate calcium intake should be assured for persons who are taking potassium thiocyanate. Use of this drug may be contraindicated in the presence of bone malacia, such as senile osteoporosis or osteitis deformans; likewise its use may be inadvisable in the presence of fracture, not as regards union of the fracture but because any interference with calcium metabolism in the presence of increased calcium requirement might precipitate osteoporosis.

BLOOD VOLUME CHANGES AND AVAILABLE (THIOCYANATE) FLUID SPACE IN SOLDIERS WITH CHRONIC WOUND INFECTIONS AND ASSOCIATED NUTRITIONAL DEPLETION

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During World Wars I and II, studies correlating blood volume changes with the clinical signs of shock have been carried out. Few data are available concerning the changes in blood volume which occur in individuals with chronic wound infections and extensive losses in body weight, conditions often present weeks or months after severe trauma. Recent observations by Lyons[16] have led him to conclude that such patients show: (*a*) an increased interstitial (thiocyanate) fluid volume as compared with standards for their weight before injury, (*b*) a reduction of 1.5 to 2 liters in blood volume, (*c*) a deficit in the total amount of circulating hemoglobin. Gregersen[11] has observed similar cases in which the blood volume, evaluated in terms of normal body weight, was reduced 30 to 40%.

Our experience with traumatically injured soldiers who had chronic wound infections and considerable weight loss has indicated that such patients are particularly susceptible to shock after surgical procedures. Although many of these individuals have weight losses ranging between 15 and 30 kilos, their concentrations of plasma proteins and red cells differ very little from those shown by normal man. It is apparent that the latter values are not good indices of changes in the blood volume. In 1945 the present investigation was undertaken to measure the blood volume and to determine the state of dehydration in battle casualties with chronic wound infections. It was hoped that such a study might result in a better guide for therapeutic management than that afforded by routine laboratory methods.

Method and Procedure. A group of 27 wounded male soldiers varying between 19 and 40 (average 22.2) years of age was studied. All had undergone a period of strenuous military training and were regarded as normal prior to injury. Several were paratroopers, the rest being infantrymen. The results obtained upon these patients are compared with those obtained from a group of 34 German prisoners of war whose ages ranged between 16 and 43 (average 26.5) years of age. These volunteers who were selected at random had been fed adequate American rations for over a year and were engaged in manual labor.

In all instances the determinations were made before breakfast with the subjects in a basal, post-absorptive state. Most of the patients were confined to their beds. In any event, the measurements were made under basal, post-absorptive conditions. None of the patients showed any evidence of cardiac failure or of peripheral vascular collapse. All were conscious, mentally coherent, and able to respond satisfactorily to questions. In the case of recent arrivals in the hospital, intravenous therapeutic procedures were postponed until some of our tests were completed. The entire group of normal subjects, and 15 of the 27 patients (Cases A1 to A15, Tables 2 and 3) were accurately weighed on a calibrated platform scale. The other 12 patients (Cases B16 to B27) were included although their clinical conditions or partial immobilization in fixed plaster casts prevented

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accurate body weight determinations. In these the weight loss was estimated, and only those in which the loss was judged to be 15 kilos or more were studied.

The plasma volume was measured with the blue dye, T-1824, using the simplified method described by Gregersen[12] in which the dye concentration is estimated with the Nickerson Decade Photometer. In most of the patients, the arms and forearms were repeatedly soaked with warm, wet towels, and both hands were immersed in warm water for 10 minutes prior to venipuncture. The skin at the site of venipuncture was first infiltrated with 1% novocaine containing no epinephrine. In individual instances in which both antecubital veins were not available, one of the external jugular veins was used.

In patients in whom both plasma volume and available (thiocyanate) fluid volume determinations were made, the procedure was carried out as follows:[13] after anesthetizing the skin, one antecubital vein was punctured with a 19-gauge needle and a dye-free blood sample was collected without stasis. Six cc. of blood were now collected in each of 2 tubes, one containing a small amount of mineral oil, the other coated with a thin film of dry heparin.* Through the same needle, 15 cc. of a 5% solution of sterile sodium thiocyanate† in an accurately calibrated syringe, were injected. The syringe was washed 3 times by drawing and reinjecting blood. Through the same needle 5 cc. of T-1824 from ampoules‡ containing a known amount of dye solution of known concentration were injected, the syringe rinsed as described above, the time recorded, and the needle withdrawn. Exactly 10 minutes later a sample (6 cc.) of blood was withdrawn without stasis from the opposite antecubital vein and transferred to a 10-cc. tube containing heparin for determination of the plasma concentration of the dye. In 5 patients 6 cc. of blood were withdrawn without stasis at the end of 1, 2, and 3 hours and transferred to test tubes containing a small amount of mineral oil for the estimation of the serum concentration of sodium thiocyanate. Since the 2 and 3 hour samples did not differ from the 1 hour sample, the practice of drawing blood at the end of 2 and 3 hours was discontinued in the subsequent measurements. The heparinized blood (dye-free and 10 minute dye-tinged) samples were transferred to separate 4-cc. hematocrit tubes (10 mm. x 8 mm.)‖ and centrifuged for 30 minutes at 3000 r.p.m. The hematocrit values obtained in the dye-free and dye-tinged samples agreed to within 2%, affording confirmatory evidence of the absence of hemostasis in the collection of the blood. Hemolyzed samples were discarded, the plasma volume determination being repeated. After centrifugation the plasma of the 10 minute dye-tinged sample was read at 624 mμ. in 10 mm. cells against a dye-free blank with the Nickerson Decade Photometer. The optical density was noted, and the plasma volume read from the appropriate table.[12] The total blood volume was calculated from the equation:

$$\text{Total blood volume (cc.)} = \frac{\text{Plasma volume (cc.)}}{\text{1-hematocrit}}$$

The plasma volume determined from a single 10 minute point is, on the average, 3.2% greater than that measured from time-concentration curves.[19] For our purposes, the method is adequate because the error is slight compared with the changes in plasma volume measured on our patients. Moreover, we have compared our results on injured soldiers with values obtained by the same method on the control group.

The available (thiocyanate) fluid volume was measured as follows: To 2 cc. of serum from the 1 hour blood sample, 2 cc. of 2% trichloracetic acid were added. The resultant mixture was centrifuged at 3000 r.p.m. for 10 minutes, and to 2 cc. of the supernatant

* Five mgs. of pure heparin were dissolved in a small amount of distilled water in 10 cc. test tubes which were placed in a tilted position in a dessicator and periodically rotated until dry, so that a thin film of heparin was left coated upon the inner surface of each tube.

† The ampoules of 5% sterile sodium thiocyanate were supplied by the Warner Institute of Therapeutic Research, N. Y. C.

‡ The ampoules containing exactly 5 cc. of a carefully standardized solution of T-1824 were supplied by the Warner Institute of Therapeutic Research, N. Y. C. This was rechecked for accuracy of standardization according to procedure outlined by Gregersen.[12]

‖ MacAlaster Bicknell Co., Cambridge, Mass.

fluid were added 2 cc. of ferric nitrate solution. The red color thus developed was read immediately in a Coleman Junior Spectrophotometer set at 460 mμ. against the similarly treated control sample. The available (thiocyanate) fluid volume was calculated from the formula given below.†

The plasma protein concentrations were determined by the falling drop method of Barbour and Hamilton,[1] using the formula of Weech *et al.*[30] The hemoglobin determinations were made with the Fisher electrohemometer.[18]

The total blood volume calculated from the plasma volume (dye method) and the venous hematocrit has been regarded as erroneous by some investigators,[7,25,26] owing to differences between the values for the venous hematocrit reading and that found for the total body hematocrit. This difference is said to result from the unequal distribution of red cells in the various vessels of the vascular system. However, the report of Hopper, Tabor, and Winkler[14] shows that the average blood volumes measured simultaneously by dye and carbon monoxide methods in man and dog are almost identical. Further evidence that the central hematocrit reading does not differ markedly from the total body hematocrit has been obtained recently by Root, Roughton, and Gregersen[23] who also obtained close agreement between the blood volumes obtained by the simultaneous measurement with dye and carbon monoxide. On the basis of this evidence, we feel that the total blood volume can be calculated from the plasma volume and the venous hematocrit reading.

The available (thiocyanate) fluid volumes were calculated from the uncorrected formula shown above. Although this ignores the fact that thiocyanate diffuses into the erythrocytes and that it is present in serum partly in bound form,[15] we feel that useful results can be obtained with this method.

Results. Normal Group. The results obtained on 34 normal individuals are shown in Table 1. These include determinations of plasma volume, available thiocyanate fluid volume, venous hematocrit reading, hemoglobin value, plasma protein concentration, and calculations of total blood volume, red cell volume, total circulating hemoglobin, and total circulating plasma proteins. Distribution curves based on the individual values for the plasma volume and blood volume expressed as cc. per cm. height, cc. per kg. body weight, and as liters per sq. m. body surface indicate, in agreement with the results of Rowntree and Brown,[24] that although the plasma and blood volume correlate well with height and weight, the correlation with body surface is better. The mean values for the various measurements, with their respective standards of deviation and coefficients of variation are given at the bottom of Table 1. Distribution curves of the various measurements are shown in Figure 1.

The plasma volume of the 34 normal males ranged from 1.31 to 2.20 liters per sq. m., the mean being 1.76 liters per sq. m. (S.D. ± 0.20). In terms of cc. per kg. body weight, the values are 32.4 to 56.9, mean 45.3 (S.D. ± 5.5). The total blood volume showed a greater range, varying from 2.14 to 4.10 liters per sq. m. of body surface, The mean value for the entire group was 3.12 liters per sq. m. (S. D. ± 0.36). Expressed in terms of cc. per Kg. of body weight, these values become 57.5 to 106.0, mean 80.1 (S. D. ± 16.5).

The blood volume and other measurements shown in Table 1 agree well with similar determinations reported by other investigators.[9,20,29]

In agreement with the report by Crandall and Anderson,[7] our results indicate that the available (thiocyanate) fluid volume correlates better with surface area than with body weight. The volumes for the 34 normal subjects (Fig. 1, *G*) ranged from 8.1 to 11 liters with a mean value of 9.8 liters per sq. m. (S.D. ± 0.61).

Wounded Soldiers. The revised diagnoses of the 27 patients studied are shown in Table 2. This table also includes a statement of the nature of the wound sustained. The individual patients have been assigned the same symbol in Table 2 as in Table 3. Since from 7 to 24 weeks had elapsed between the time of injury

† $\text{Available fluid in liters} = \frac{\text{Total amount of NaCNS injected in gm.} \times 100}{\text{mg. NaCNS/100 cc. serum after equilibrium is established.}}$

TABLE 1.—BODY FLUID DATA OBTAINED FROM 34 NORMAL ADULT MALES

No.	Age	Height (cm.)	Weight (kg.)	Surface area (M^2)	Plasma volume			Blood volume			RBC volume			Plasma prot.		Hemoglobin		Available fluid			Hematocrit (%)
					Total L	Cc./kg	L/M^2	Total L	Cc./kg	L/M^2	Total L	Cc./kg	L/M^2	Conc. (gm.%)	Total circ.(gm.)	Gm.%	Total circ.(gm.)	Total L	Cc./kg	L/M^2	
1	19	174	63.5	1.76	2.80	44.1	1.59	5.05	79.5	2.86	2.25	35.4	1.28	6.8	190	15.8	794	16.7	263	9.5	44.5
2	26	171	69.0	1.80	3.00	43.6	1.66	5.26	71.2	2.90	2.26	32.8	1.25	7.2	216	14.5	763	17.4	252	9.7	43
3	20	176	68.2	1.83	3.60	52.8	1.96	6.43	94.2	3.51	2.83	41.5	1.54	6.5	234	15.6	1007	17.2	252	9.4	44
4	31	174	71.4	1.84	3.70	51.8	2.01	6.25	87.5	3.40	2.55	35.7	1.38	6.6	244	15.1	944	19.7	276	10.7	44
5	23	178	69.0	1.85	3.00	43.5	1.63	5.34	77.5	2.90	2.34	33.8	1.26	7.3	234	15.7	838	15.0	218	8.1	44
6	24	174	72.7	1.86	3.05	42.1	1.64	5.26	72.5	2.82	2.20	30.3	1.18	6.2	189	15.2	800	18.3	253	9.8	42
7	33	173	76.4	1.89	3.70	48.5	1.96	6.30	82.5	3.40	2.60	34.0	1.37	6.9	255	14.3	900	18.4	241	9.7	41
8	27	176	75.0	1.91	3.10	41.3	1.60	5.50	70.3	2.90	2.43	32.5	1.27	6.7	208	15.3	846	18.0	240	9.4	44
9	24	180	75.5	1.94	4.30	56.9	2.20	8.00	106.0	4.10	3.70	49.0	1.90	6.7	288	15.3	1224	21.0	278	10.8	46
10	21	178	77.2	1.94	3.90	50.5	2.00	6.72	87.0	3.46	2.82	36.5	1.45	7.2	280	14.9	1000	19.7	255	10.1	42
11	19	178	75.7	1.94	3.60	47.5	1.86	6.46	85.0	3.33	2.86	37.8	1.48	6.4	230	15.6	1000	18.2	240	9.4	44.5
12	20	181	76.4	1.96	2.80	36.7	1.40	5.70	74.5	2.90	2.90	38.0	1.48	7.2	202	16.7	954	18.6	244	9.5	51
13	24	176	81.0	1.97	3.60	44.5	1.80	6.05	74.9	3.10	2.45	30.4	1.24	6.7	240	13.6	820	18.4	228	9.3	40.5
14	23	174	83.0	1.97	3.70	44.6	1.87	6.80	82.0	3.40	3.10	37.5	1.57	7.1	263	15.0	1000	19.8	240	10.0	45.5
15	20	171	63.5	1.74	3.15	49.5	1.80	5.23	82.3	3.00	2.08	32.8	1.20	7.4	233	14.8	774	17.2	271	9.8	43
16	33	156	52.0	1.48	2.70	52.0	1.83	4.43	85.1	2.90	1.73	33.2	1.17	6.6	178	14.9	646	14.5	278	9.8	39
17	32	156	58.2	1.55	2.80	48.2	1.80	5.00	85.9	3.20	2.20	37.7	1.42	7.2	202	15.9	795	15.3	262	9.9	44
18	39	154	61.7	1.59	3.40	55.2	2.13	5.85	95.0	3.60	2.45	39.7	1.54	6.9	235	15.0	878	15.4	250	9.7	42
19	23	157	60.5	1.60	2.85	47.2	1.80	5.03	83.4	3.14	2.18	36.0	1.36	6.8	194	13.6	684	15.8	261	9.9	43
20	16	164	60.5	1.65	2.80	46.3	1.70	4.59	75.8	2.80	1.79	29.6	1.09	7.1	199	14.0	642	16.0	265	9.7	39
21	45	165	62.7	1.68	2.20	35.1	1.31	3.60	57.5	2.14	1.40	22.4	0.83	6.9	152	14.1	508	16.0	216	9.5	42
22	25	164	63.5	1.68	2.60	41.0	1.55	4.56	72.0	2.70	1.96	30.9	1.17	7.0	182	14.8	675	16.7	263	9.9	43
23	20	168	61.8	1.69	2.90	47.0	1.70	5.00	81.0	2.95	2.10	34.0	1.24	6.9	200	14.8	740	15.3	248	9.1	42
24	29	169	61.8	1.70	2.90	47.0	1.70	5.47	88.6	3.22	2.57	41.7	1.51	7.2	209	15.1	826	16.0	260	9.4	47
25	39	157	69.0	1.70	3.10	44.8	1.82	6.00	87.0	3.52	2.90	42.0	1.70	6.5	202	16.0	960	19.0	276	11.0	48
26	20	166	64.5	1.71	3.40	52.8	2.00	5.81	90.1	3.40	2.41	37.4	1.41	7.0	238	13.9	808	17.7	275	10.3	42
27	43	162	69.5	1.74	2.25	32.4	1.30	4.24	61.0	2.40	1.99	28.7	1.14	7.0	158	15.9	674	15.5	222	9.0	47
28	21	169	72.2	1.82	3.40	47.1	1.87	6.30	87.0	3.46	2.90	40.2	1.59	6.6	224	14.8	932	19.4	268	10.5	46
29	34	184	83.6	1.90	3.30	39.5	1.74	5.50	66.0	2.90	2.20	26.4	1.16	6.5	215	14.3	787	20.2	241	10.6	40
30	19	170	87.2	1.98	3.70	42.4	1.80	6.50	74.5	3.30	2.80	32.1	1.40	6.7	250	15.1	980	19.7	226	10.0	43
31	23	170	88.5	1.99	3.75	42.4	1.88	6.58	74.3	3.30	2.83	33.3	1.40	6.9	260	13.6	895	19.3	218	9.8	43
32	23	168	67.2	1.75	2.90	43.2	1.70	5.09	75.5	2.93	2.19	32.6	1.25	7.3	210	13.4	680	17.7	262	10.1	43
33	21	166	72.2	1.80	3.15	43.6	1.75	5.72	79.2	3.18	2.57	37.4	1.43	7.6	239	16.1	920	18.9	261	10.5	45
34	41	115	101.0	2.30	3.75	37.1	1.63	6.80	67.3	2.96	3.06	30.3	1.33	7.4	278	16.1	1096	19.3	192	8.4	45
Total	900	5696	2415.1	61.50	108.85	1542.2	59.98	192.40	2713.0	105.94	83.58	1183.6	45.99	235.5	7531	508.8	28 790	601.3	8495	332.3	1481
Mean	26.5	168	71.0	1.81	3.20	45.3	1.76	5.66	80.1	3.12	2.46	34.8	1.35	6.9	221	15.0	847	17.7	249	9.8	43.5
Standard deviation						5.5	20		16.5	365		5.1	0.195	.35		0.8			22	0.61	2.5
Coefficient of variation						12.1	11.3		20.6	11.7		14.6	14.5	5.1		5.3			8.9	6.1	5.7
Mean (gm./kg)															3.12		11.5				
Standard deviation															.39		1.6				
Coefficient of variation															12.4		13.4				

Fig. [illegible] comparison values [illegible]

No. of Cases

No. of Cases

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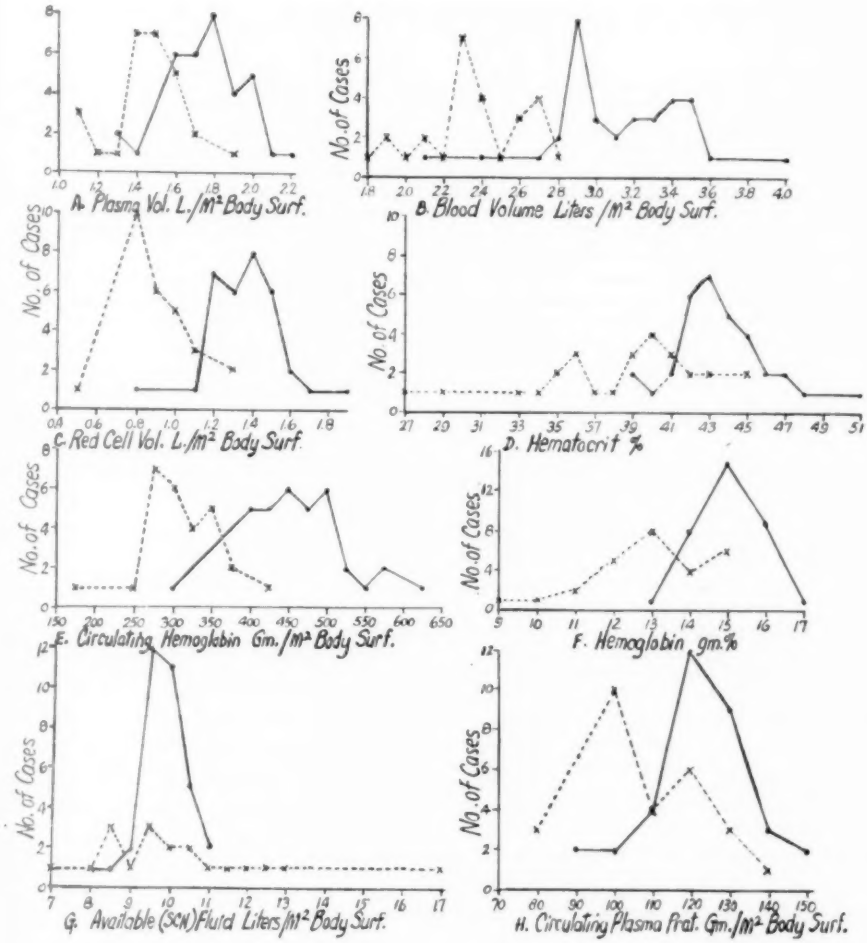
A. Plasma Vol. L./M² Body Surf.
B. Blood Volume Liters/M² Body Surf.
C. Red Cell Vol. L./M² Body Surf.
D. Hematocrit %
E. Circulating Hemoglobin Gm./M² Body Surf.
F. Hemoglobin gm.%
G. Available (SCN) Fluid Liters/M² Body Surf.
H. Circulating Plasma Prot. Gm./M² Body Surf.
No. of Cases

FIG. 1.—Distribution curves from body fluid data obtained from the 27 wounded soldiers studied compared with corresponding data from 34 normal male controls. The closed circles represent individual values in the normal control group. The crosses represent individual values in the patient group.

and the time at which our observations were made (see Table 2), many of our patients had recovered from the effects of some of the initial wounds. Others had developed various complications, some of which were remotely associated with and of graver import than the initial injury. Hence, the original diagnoses listed in such accompanying tags, records, and charts as were available to us have been revised to conform with the clinical status of each patient at the time he was studied (Table 3).

Practically all of the wounds were of the complex polymicrobial type similar to those described by Lyons[16] and Pulaski and Sprinz.[22] The extent and character of the anatomical involvement were also extremely varied (see Table 2). All of our patients had had surgical attention, had received either sulfonamides, or penicillin, or both, as well as variable amounts of plasma or whole blood at different medical installations. Accurate data concerning previous transfusions or other parenteral

TABLE 2.—DIAGNOSES OF THE 27 WOUNDED SOLDIERS REPORTED IN THIS STUDY (EACH SYMBOL IN TABLES 2 AND 3 REFERS TO THE SAME PATIENT)

Case	
A1	Wounded 21 weeks previously, struck by mast of ship attacked by bombers; simple compression fracture of 1st lumbar vertebra; no cord injury; traumatic subdural cyst, cauda equina (healed); multiple, large decubitus ulcers; acute bilateral pyelonephritis; chronic cystitis (suprapubic cystotomy).
A2	Accidentally injured 21 weeks previously with resulting compression of the cauda equina, and paraplegia secondary to fracture and compression of 1st lumbar vertebra; compound linear fracture right frontal bone, healed; simple fracture, 5, 6 right ribs, healed; contusion right kidney, healed; nephrolithiasis with acute pyelonephritis, bilateral; chronic cystitis (suprapubic cystotomy); multiple decubitus ulcers.
A3	Wounded by shell fragments 20 weeks previously; fracture, compound, comminuted of 2, 3, 4, 5 left metatarsals with no evidence of callus formation after 4 months; fracture, simple, right humerus, healing; partial paraplegia secondary to comminuted fracture (healed), 4th lumbar vertebra; chronic cystitis (suprapubic cystotomy); multiple, large granulating decubitus ulcers.
A4	Wounded by bullet 17 weeks previously; penetrating bullet wound right chest; fracture dislocation 4, 5, 6 thoracic vertebræ; severe partial traumatic myelitis and paraplegia with sensory loss up to D9; chronic empyema (treated by open drainage) and bronchopleural fistula right chest; chronic cystitis (suprapubic cystotomy); multiple decubitus ulcers.
A5	Wounded 10 weeks previously by shrapnel; perforating wound severe entering left flank and with exit at right flank with resultant laceration of cecum (cecotomy and appendectomy performed on date of wound); incomplete compound comminuted fracture left ilium with chronic osteomyelitis; fecal fistula left flank.
A6	Wounded 11 weeks previously by shrapnel; wound, suppurating, soft tissues right gluteal region with draining abscess, deep; no fracture.
A7	Wounded by shrapnel 14 weeks previously; suppurating wound soft tissues right deltoid region; no bone injury; postdiphtheritic polyneuritis and generalized motor paralysis.
A8	Wounded by shrapnel 9 weeks previously; multiple penetrating wounds (healed) left foot, leg, thigh and gluteal region, right thigh, and left forearm; compound complete fracture (healing) left radius; paralysis left ulnar nerve; purulent drainage from fistulous tract, right pararectal region, secondary to lacerated wound with large perforation posterior rectal wall.
A9	Wounded 22½ weeks previously by shrapnel; chronic osteomyelitis, shaft right femur, secondary to compound comminuted fracture; large chronic suppurating wound right thigh.
A10	Wounded by shrapnel 8 weeks previously; chronic, deep, draining abscess left thigh extending into Hunter's canal; paralysis left femoral nerve; simple, incomplete fracture neck of left femur; simple comminuted fractures 8, 9 right ribs; suppurating wound right flank (5 by 10 cm.).
A11	Wounded by bullet 10½ weeks previously, perforating wound abdomen with lacerations of colon, mesentery and jejunum (colostomy present, intestinal lacerations healed); comminuted fracture 4th, 5th lumbar vertebræ; traumatic myelitis, with paraplegia; chronic retroperitoneal abscess; osteomyelitis, ilium, left; acute pyelonephritis, right; chronic cystitis (suprapubic cystotomy).
A12	Wounded by bullet 15 weeks previously; perforating wound abdomen with 4 perforations of ilium (healed); suppurating wounds left flank, left leg; draining psoas abscess, left.
A13	Wounded by shrapnel 21 weeks previously; penetrating wound right flank with shell fragment lodged at 4th lumbar vertebra; fecal fistula right flank to ascending colon; draining subdiaphragmatic abscess, right; laceration nerve roots (4 and 5 lumbar and 1st sacral) with secondary paresis; chronic cystitis with bilateral ascending pyelonephritis.
A14	Wounded by shrapnel 19 weeks previously; penetrating wound, right gluteal region; chronic osteomyelitis, head of right femur and acetabulum secondary to compound comminuted fracture; pyoarthrosis right knee joint; fecal fistula descending colon; retroperitoneal abscess; colostomy.
A15	Wounded 14 weeks previously by bullet, left flank with perforation of stomach and injury to pancreas and kidney (all healed) except for draining sinus, left flank.
B16	Wounded 14 weeks previously by shell fragment; chronic osteomyelitis, right femur, secondary to compound comminuted fracture.
B17	Wounded by bomb fragment 12 weeks previously; perforations of stomach (healed); perforation of hepatic flexure (exteriorized as colostomy); double barrel jejunostomy done for intestinal obstruction; intra-abdominal mural abscess; perihepatitis; chronic peritonitis, bronchopneumonia; determinations done 3 weeks before death.
B18	Wounded by bomb fragments 12 weeks previously; chronic osteomyelitis (resolving) secondary to compound fracture, left tibia and fibula.

TABLE 2.—DIAGNOSES OF THE 27 WOUNDED SOLDIERS REPORTED IN THIS STUDY (EACH SYMBOL IN TABLES 2 AND 3 REFERS TO THE SAME PATIENT)—CONTINUED

Case	
B19	Wounded by mine fragments 19 weeks previously; chronic osteomyelitis, ischii, pubes, sacrum, coccyx, and necks of both femurs, secondary to compound comminuted fractures; chronic cystitis (suprapubic cystotomy); suppurating wounds, right thigh; multiple decubitus ulcers.
B20	Wounded by bullet 24 weeks previously; penetrating gunshot wound resulting in fracture, 7th cervical vertebra, with traumatic myelitis and complete paralysis below C7; clean granulating stump left leg after amputation below knee 1 month previously for gangrene; multiple large decubitus ulcers; chronic cystitis (suprapubic cystotomy) tests done 4 days before death (preceded by unexplained convulsions).
B21	Wounded 15 weeks previously by bullet; penetrating wounds, buttocks and rectum; colostomy closed; wounds granulating slowly.
B22	Accidentally injured in jeep 7 weeks ago; compound septic necrosis left knee joint and simple fracture rim of left acetabulum.
B23	Wounded by shell fragment; wounds soft tissues right leg (granulating slowly); no fracture.
B24	Wounded by shell fragment 16 weeks previously with infected wound soft tissues left leg; amputation stump right leg (healed).
B25	Wounded by shell fragments 7 weeks previously with granulating wounds right ankle, elbow and shoulder; fracture, right ankle (uniting).
B26	Wounded 12 weeks previously by shell fragment; chronic osteomyelitis right femur, tibia, fibula and cuneiform bone and 1st metacarpal secondary to multiple compound comminuted fractures.
B27	Wounded by shell fragment 19 weeks ago; chronic osteomyelitis right femur, tibia and fibula and left tibia, secondary to multiple compound comminuted fractures; suppurative arthritis and synovitis, both knee joints.

therapy were generally incomplete. Eight of our patients had paraplegia. Measurements of the height and weight before injury were available for all the patients, from which the normal surface area could be determined in the usual way.[3] The body fluid measurements have been computed in terms of liters per sq. m. of normal body surface. The results of the measurements of plasma, total blood, red cell, and available fluid volumes are shown in Table 3. Hematocrit readings, and hemoglobin and plasma concentration also appear in this table. For comparison with the results obtained in the normal group, the frequency distribution curves of the body fluid measurements of the patient group are presented in Figure 1.

We have compared the mean total volume measurements of plasma, total blood, red cells, and available thiocyanate fluid of the patient group with estimated normal values obtained by multiplying the mean normal surface area of the patients by the mean volume per sq. m. of body surface measured in the normal group. The mean hematocrit readings, and hemoglobin and plasma protein concentrations of the 27 patients are compared with the corresponding mean value of the 34 normal males. Their values are given in Table 4. In column 5 of this table, the mean changes from the estimated normal appear, and in column 6 is given the percentage increase or decrease from the normal. The statistical significance of the change (column 7) is expressed by dividing the values of column 5 by the standard error.

Blood Volume. The average plasma volume of the 27 patients was 2760 cc. which is 580 cc. or 17.3% less than the mean estimated normal value. Similarly based comparisons for the total blood and red cell volumes of these patients showed an even greater decrease for the mean total blood volume amounting to 1430 cc. (24.2%), and a reduction in the mean red cell volume equal to 815 cc. (31.6%). The reduction in total blood volume is the result of decreases in both plasma volume and red cell volume, but the decrease in the latter is 1.8 times that shown by the former. The average hematocrit of our patients was 38.4%, which is 11.7% less than the mean hematocrit for the control group. The reduction in the mean red cell

TABLE 3.—BODY FLUID DATA OBTAINED FROM 27 SOLDIERS WITH CHRONIC WOUND INFECTION, NUTRITIONAL DEPLETION, AND WEIGHT LOSS (EACH SYMBOL IN TABLES 2 AND 3 REFERS TO THE SAME PATIENT)

Case	Age	Height (cm.)	Weight			Surface area		Plasma volume		Blood volume		RBC volume		Plasma protein		Hemoglobin		Available fluid			Hematocrit (%)
			Normal (kg.)	Present (kg.)	Loss (kg.)	Normal (M^2)	Present (M^2)	Total L.	Normal S.A. (L/M^2)	Total L.	Normal S.A. (L/M^2)	Total L.	Normal S.A. (L/M^2)	Gm. %	Total circ. (gm.)	Gm. %	Total circ. (gm.)	Total L.	Normal S.A. (L/M^2)	Present S.A. (L/M^2)	
A1	21	170	63.5	31.5	32.0	1.74	1.28	1.90	1.09	3.22	1.85	1.32	0.76	8.8	167	14.7	473	18.0	10.3	14.1	41.0
A2	24	170	57.0	40.0	17.0	1.65	1.42	2.20	1.33	3.44	2.08	1.24	0.75	8.0	176	12.7	437	13.8	8.4	9.7	36.0
A3	19	179	64.5	38.5	26.0	1.80	1.44	2.60	1.44	4.16	2.30	1.59	0.88	6.8	173	14.7	611	16.0	8.9	11.0	37.5
A4	31	177	66.0	44.5	21.5	1.81	1.53	2.80	1.55	4.24	2.34	1.44	0.80	7.0	195	12.1	513	23.0	12.7	14.0	34.0
A5	19	177	82.0	53.5	28.5	1.98	1.66	2.90	1.46	4.50	2.26	1.60	0.81	7.1	206	12.3	660	23.0	11.6	14.0	35.5
A6	21	180	78.5	52.5	26.0	2.02	1.70	2.80	1.39	4.75	2.36	1.95	0.97	7.2	202	13.0	617	20.2	10.0	11.8	41.0
A7	23	180	70.5	56.0	14.5	1.89	1.71	2.30	1.22	3.90	2.06	1.60	0.85	7.9	182	14.5	566	17.6	9.3	10.3	41.0
A8	26	182	72.5	48.0	24.5	1.92	1.61	2.10	1.09	3.70	1.93	1.60	0.84	7.3	153	15.2	562	13.3	7.0	8.3	43.0
A9	20	177	79.5	59.5	20.0	1.97	1.73	2.65	1.35	4.31	2.18	1.66	0.85	7.7	204	13.2	569	16.2	8.3	9.4	38.5
A10	28	184	77.0	55.0	22.0	1.99	1.71	3.75	1.88	5.28	2.66	1.53	0.77	7.2	270	9.9	523	16.2	8.2	9.4	29.0
A11	27	179	84.0	56.0	28.0	2.02	1.69	2.80	1.39	5.09	2.52	2.29	1.13	5.9	165	14.1	718	21.2	10.5	12.5	45.0
A12	26	175	68.0	45.0	23.0	1.82	1.53	2.70	1.48	4.12	2.26	1.42	0.78	8.0	216	10.6	437	18.4	10.1	12.0	34.5
A13	32	170	70.5	43.0	27.5	1.80	1.46	3.10	1.72	4.92	2.72	1.82	1.02	7.5	232	12.0	560				37.0
A14	23	180	78.5	48.5	30.0	1.97	1.61	2.90	1.47	3.97	2.02	1.07	0.54	8.2	239	9.2	365				27.0
A15	19	172	70.5	53.0	17.5	1.83	1.62	2.60	1.42	4.30	2.34	1.70	0.93	8.2	213	13.2	568				39.0
B16	25	172	59.1	43.2*	15.9*	1.69	1.48	1.80	1.06	3.10	1.83	1.30	0.77	7.4	135	15.1	468	14.7	8.7	9.9	42.0
B17	32	172	63.6	43.2*	20.4*	1.75	1.55	2.80	1.60	4.79	2.74	1.99	1.14	6.0	168	13.6	651	22.7	13.0	14.6	41.5
B18	32	175	69.5	50.0*	19.5*	1.82	1.60	2.80	1.54	5.09	2.80	2.29	1.25	7.5	210	15.2	775				45.0
B19	31	177	75.0	50.0*	25.0*	1.91	1.60	3.00	1.57	4.88	2.56	1.88	0.99	7.3	219	13.3	649	22.5	11.8	14.0	38.5
B20	28	175	81.8	50.0*	31.8*	1.96	1.60	3.00	1.53	4.61	2.34	1.61	0.82	6.8	204	11.2	515	33.2	17.0	20.7	35.0
B21	20	177	75.1	54.7*	20.4*	1.91	1.62	2.70	1.41	4.50	2.36	1.80	0.94	7.2	195	14.0	730	18.0	9.5	11.1	40.0
B22	26	175	75.1	54.7*	20.4*	1.90	1.65	3.10	1.63	4.59	2.42	1.49	0.79	7.4	230	11.5	528	17.9	9.4	10.9	33.0
B23	20	177	70.6	54.7*	15.9*	1.87	1.68	2.75	1.47	4.58	2.44	1.83	0.98	8.8	242	13.9	637				40.0
B24	22	180	77.4	54.7*	22.7*	1.95	1.69	3.30	1.69	5.15	2.64	1.85	0.95	7.5	248	12.2	630				36.0
B25	23	182	75.0	59.1*	15.9*	1.95	1.76	2.85	1.46	5.00	2.56	2.15	1.10	7.6	216	12.9	645				43.0
B26	20	190	93.8	63.8*	30.0*	2.24	1.81	3.05	1.36	5.04	2.26	1.99	0.89	7.5	229	13.2	665				39.5
B27	40	182	86.5	68.3*	18.2*	2.04	1.87	3.30	1.62	5.50	2.70	2.70	1.32	7.1	234	12.5	688	22.7	11.1	12.0	40.0
Total		4786	1985.0	1370.9	614.1	51.27	43.68	74.55	39.22	120.73	63.53	46.71	24.62	200.9	5523	350.0	15,810	368.6	195.8	229.7	1032.5
Mean		178	73.5	50.8	22.8	1.92	1.62	2.76	1.45	4.49	2.36	1.75	0.91	7.4	205	15.0	585	19.4	10.3	12.1	38.4
Standard deviation										0.61		0.39		0.7	31.4	1.5	97.5	4.5			4.3
Coefficient of variation								0.42		13.6		22.3		8.9	15.3	11.6	16.7	23.2			11.3

* Estimated weight.

S. A. = Surface area.

TABLE 4.—MEAN BODY FLUID MEASUREMENTS FOR THE WOUNDED GROUP REPORTED AND THE RESPECTIVE CHANGES FROM THE ESTIMATED MEAN NORMAL BODY FLUID VALUES FOR THIS GROUP

Measurement	No. of cases	Mean measurement in wounded group studied	Mean estimated normal values*	Mean change in wounded group	Change in wounded group (%)	Difference divided by standard error
Plasma volume	27	2760 cc.	3340 cc.*	−580 cc.	−17.3	6.3
Blood volume	27	4490 cc.	5920 cc.*	−1430 cc.	−24.2	5.6
RBC volume	27	1750 cc.	2565 cc.*	−815 cc.	−31.6	10.0
Available fluid volume	19	19,400 cc.	18,500 cc.*	+900 cc.	+4.9	0.72
Circulating hemoglobin	27	585 gm.	890 gm.*	−305 gm.	−34.2	14.0
Circulating plasma protein	27	205 gm.	235 gm.*	−30 gm.	−12.8	4.8
Hematocrit	27	38.4%	43.5%†	−5.1%	−11.7	6.0
Hemoglobin	27	13.0 gm.%	15.0 gm.% †	−2.0	−13.3	6.9
Plasma protein	27	7.4 gm.%	6.9 gm.%†	+0.5	+5.0	3.3

* The estimated value is obtained by multiplying the mean normal surface area of the cases studied by the corresponding mean normal measurement per sq. m. body surface obtained from the data on the 34 normal males in Table 1.

† Mean normal values observed in the 34 normal males.

volume is therefore 2.7 times the decrease shown by the mean hematocrit. The mean hemoglobin concentration of the patient group was 13 gm. per 100 cc., or 13.3% less than the value for the control group. The mean circulating hemoglobin in our patients was 585 gm. This value is 305 gm. or 34.2% less than the mean estimated normal value. The reduction in the mean circulating hemoglobin is therefore 2.6 times that shown by the reduction in the mean hemoglobin concentration. The values in Table 3 (column 7) indicate that the above observations are statistically significant.

Available (Thiocyanate) Fluid Volume. Available (thiocyanate) fluid volume measurements were made in 19 of the 27 patients studied (Table 3). The volumes obtained when based on the normal surface area of these patients ranged from 7 to 17 liters per sq. m. In 4 cases, the volumes observed were more than 2 standard deviations below the mean normal value. The lowest value was observed in Case A8 whose available (thiocyanate) fluid volume was 7 liters per sq. m. In 6 cases, the available (thiocyanate) fluid volume per sq. m. was more than 2 standard deviations above the mean value for our normal group. Of these 6 cases, 5 showed volumes ranging from 11 to 13 liters per sq. m., while 1 case (B20) had 17 liters of available fluid per sq. m. The average total available (thiocyanate) fluid volume of the 19 patients measured was 19.4 liters which is 0.9 liters or 4.9% above the mean estimated normal value (Table 3). This increase is however, not statistically significant.

Analysis of the individual total available (thiocyanate) fluid volumes in terms of the body weight of each patient at the time this study was made show that 8 of the 19 cases measured presented volumes which were in excess of 40% of their body weight, while 4 of the 8 cases (A1, A4, B17, B20) had volumes equivalent to more than 50% of their body weight. The latter were extremely ill, febrile, and cachetic, and the total available (thiocyanate) fluid volumes observed in them were markedly elevated in comparison with the mean available (thiocyanate) fluid volume found in our normal controls which was equivalent to 25% of the body weight. The measurements in Cases A1, B17 and B20 were made 11, 22 and 3 days prior to death respectively. Two months after the first available fluid determination, a second one was made on Case A4 following marked improvement and the establishment of a positive nitrogen balance. Although this patient had gained only 3.5 kg., his total available (thiocyanate) fluid volume had decreased from 23 liters which was equal to 51.7% of his body weight, to 16.4 liters, a value equal to 34% of his body weight.

Circulating Plasma Proteins. Although weight losses of 14.5 to 32 kg. were observed in the 27 patients studied, the average plasma protein concentration for the entire group was 7.4 gm. per 100 cc.

(S.D. ± 0.7) (Table 3) which is 5% more than the mean plasma protein concentration for the control group. Plasma protein values below 6.5 gm. per 100 cc. were observed in 2 cases, while values above 8 gm. per 100 cc. or more were present in 6 cases. The average total circulating plasma protein in the 27 patients was 205 gm. (S.D. ± 31.4) which is 30 gm., or 12.8% less than the mean estimated normal value. The linear correlation of the plasma volume to total circulating plasma protein in our normal controls is shown in Figure 2. From the same figure, it is apparent that values for the plasma volume in our 27 patients are generally below normal, although they tend to be disposed along the same axis of drift displayed by the plotted values for the normal control group.

Discussion. Patients with chronic wound infections, nutritional depletion, and anemia show a 24.2% reduction in blood volume as compared with the estimated mean normal value. This is the result of a 17.3% decrease in plasma volume and a 31.6% reduction in red cell volume. The condition may be described as an oligocythemic hypovolemia. According to Stead and Ebert[27] the blood volume of normal subjects with a relatively constant arterial pressure is related to the amount of circulating plasma proteins. Our observations show a linear correlation between the plasma volume and the circulating plasma protein in the normal and patient groups (Fig. 2). Madden and Whipple[17] report that the plasma proteins are in dynamic equilibrium with the tissue proteins. This seems likely in our patient group, for the reduction in weight must have occurred at the expense of fat and protein. Negative nitrogen balances were observed in several cases on whom such studies were done. The metabolic status of these individuals is that of semi-starvation. It is of primary importance that proper attention be given to insure an adequate protein intake in these patients so that positive nitrogen equilibrium may be restored.

Although the patients with chronic

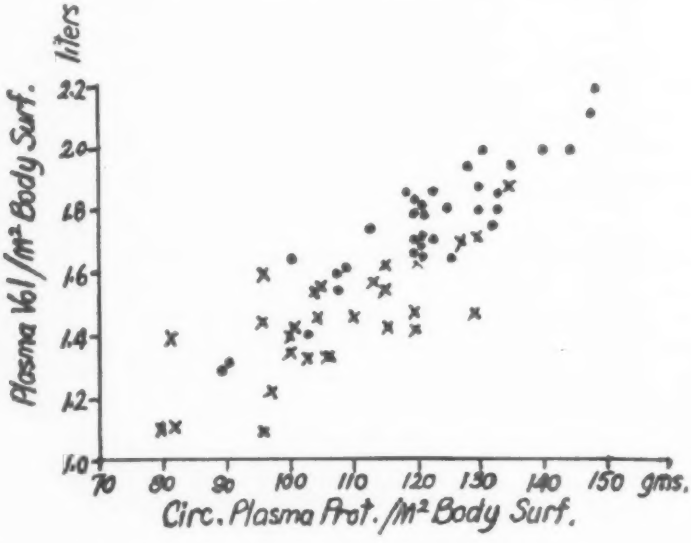

FIG. 2.—Plasma volume plotted against circulating plasma proteins per square meter normal body surface in the 27 wounded soldiers studied and in 34 normal male controls. The closed circles represent individual values in the normal control group. The crosses represent individual values in the patient group.

wound infections and pronounced weight loss studied by us showed a slightly elevated mean plasma protein concentration, their mean total circulating plasma protein was actually 12.8% less than the estimated mean normal value. Sole reliance on plasma protein levels as indices of the plasma volume, circulating plasma proteins, and nutritional status of such patients may therefore be quite misleading. Two of our patients had plasma protein values below 6.5 gm. per 100 cc., and 6 patients had levels of 8 gm. or more. Evidence of dehydration in the latter was absent. Available (thiocyanate) fluid volumes measured in 3 of these patients were normal. While data on the plasma protein fractions in our patients are incomplete, the elevated plasma protein values observed are, in all likelihood, the result of an elevated globulin fraction and concomitantly lowered albumin fraction as has been noted in chronic suppurative infections.[2,4] It is highly probable that the reduction in plasma volume is largely the result of a lowered osmotic efficiency of the plasma secondary to a reduction in circulating plasma albumin.

The changes in the hematocrit in our 27 patients failed to indicate reliably the extent of change in the circulating red cell volume, for the average reduction in the latter was 2.7 times that shown by the former. Our observations are in agreement with those of Hopper *et al.*[14] that the dye method indicates changes in the plasma volume more readily than changes in the serum protein concentration or hematocrit.

Similarly, changes in the hemoglobin concentration were poor indices of the degree of oligochromemia in our patients, for the mean reduction in their circulating hemoglobin was 2.6 times the average decrease in the hemoglobin concentration. The anemias in our cases of chronic wound infection were refractory to liver extract and iron therapy. This has been attributed to a disturbance in the intermediary metabolism of iron with a secondary failure to form heme.[5] Our experience indicates that until the associated infections are cured, the only effective treatment for the anemias observed in these patients is transfusions of whole blood in adequate quantities repeated as frequently as necessary. Many of our patients would not have survived were it not for repeated transfusions given while measures to control these infections were being instituted.

Unlike healthy individuals, patients with chronic wound infections, nutritional depletion, and a lowered blood volume cannot withstand relatively minor blood losses without being predisposed to shock, peripheral vascular collapse, and anoxic complications. These events have been observed in such patients following surgical procedures. This is not surprising in view of the lowered blood volume, red cell volume, and circulating hemoglobin observed in them, and the fact that the blood loss in ordinary surgical operations may be considerable.[6,9] We concur with Lyons[16] that the maintenance of an adequate blood volume is of utmost importance in the prevention of shock and its complications in these patients. The results of this study emphasize the necessity for whole blood transfusions in adequate quantities in the preoperative and postoperative management of such cases. Our experience is in agreement with that of Coller *et al.*,[6] and Stewart and Rourke[28] that the quantity of blood loss is not reflected by changes in the hematocrit, hemoglobin, and plasma proteins prior to and immediately after an operation. Reliance on these determinations may lull the surgeon with a false sense of security when indeed shock may subsequently intervene. It is clear that in these patients the indication is adequate replacement with whole blood, for the administration of plasma neglects the desired augmentation of the circulating hemoglobin and serves to prolong and aggravate the existing anemia.

Of the 19 patients whose available (thiocyanate) volumes were measured, 8 (Cases A1, A3, A4, A5, A12, B17, B19, B20) (Table 3) showed volumes in excess

of 40% of the body weight noted at the time this study was made. The increased volumes observed in these cases are explained partly by the pronounced cellular wastage, fat loss, tissue destruction, and toxemia, and the replacement by interstitial fluid. In Cases A1, A4, B17 and B20, the available fluid volumes were respectively equivalent to 57, 51.7, 52.5, and 66.5% of the observed body weight. These figures denote exceedingly large increases which cannot be explained solely on the basis of increases in extracellular water. In Case B20, the value approximates that for total body water. By tissue analysis methods, Overman[21] has established that under certain conditions which are not entirely understood, thiocyanate ion appears to enter the tissue cells as a result of generalized permeability alterations. This offers a likely explanation for the excessive increases in available (thiocyanate) fluid volume observed in Cases A1, A4, B17 and B20, all of whom were extremely ill, febrile, cachetic, and suffered with severe chronic wound infections. Of the latter, only Case A4 survived. Excess fluid in the serous cavities and tissues were absent at autopsy in Cases A1, B17 and B20. A second available (thiocyanate) fluid determination in Case A4 made 2 months after the first measurement and following marked improvement and a gain in weight of 3.5 kg., showed that the available (thiocyanate) fluid volume had decreased from a value equal to 51.7% of the body weight to a value which was 34% of the body weight. We associate this change with the recovery phase of tissue permeability alterations occurring in this patient. It is our experience that in such patients a markedly elevated available (thiocyanate) fluid volume approaching the value for total body water is associated with a grave prognosis.

Summary. 1. Body fluid data on 34 normal young adults and 27 patients with chronic wound infections, extensive weight loss, and anemia are reported.

2. The patients studied show an average reduction in blood voume of 1430 cc. (24.2%). This is the result of decreases in plasma and red cell volumes, the decrease in the latter being 1.8 times that shown by the former. Maintenance of an adequate blood volume to prevent shock and its complications is of utmost importance in these cases. For this purpose whole blood in adequate quantities, given repeatedly if necessary, is indicated preoperatively and postoperatively.

3. The mean reduction in the red cell volume and circulating hemoglobin observed in our patients was approximately 2.5 times that shown by the respective mean changes in the hematocrit and hemoglobin concentration. Blood volume determinations offer a practical and more reliable guide in therapy.

4. The available (thiocyanate) fluid volumes were in excess of 40% of body weight in 8 of 19 patients measured, and equal to more than 50% of the body weight in 4 cases. The values in the latter are attributed to the entrance of thiocyanate ion into the cells as a result of generalized permeability alterations following prolonged nutritional depletion, pyrexia, severe chronic polymicrobial infections, and toxemia. In 1 case the available (thiocyanate) fluid volume approached the generally accepted value for total body water.

5. Close correlation between the reduced plasma volumes and circulating plasma proteins were observed in our patients. In general, the plasma protein concentrations failed to reflect the degree of change in the total circulating plasma protein and plasma volume and may, in fact, fail to indicate the direction of change in these cases. As an index of the nutritional status, the plasma protein concentration may be quite misleading. Re-establishment of positive nitrogen equilibrium by an adequate protein intake is indicated in all these patients.

6. The clinical implications of these findings are discussed.

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RADIOLOGY

UNDER THE CHARGE OF

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ROENTGENOLOGIC EVIDENCE OF BILIARY REGURGITATION: ROENTGENOLOGIC CONTRIBUTIONS TO THE DIAGNOSIS OF HYPOPARATHYROIDISM, APPENDICITIS; MESENTERIC THROMBOSIS

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1. *Roentgenographic Demonstrations of the So-Called Regurgitation Phenomenon in Jaundice.* In a series of 460 cholangiographic examinations in which diodrast (3,5-diiodo-4-pyridone-N-acetic acid diethanolamine) was used, Rigler and Mixer[10] observed 8 patients whose renal pelves were clearly outlined by the cholangiographic medium on roentgenograms made at intervals of from 5 to 30 minutes after the drug was injected into the common bile duct. Cholecystectomy had been performed on all of the patients and a drainage tube had been inserted into the biliary ducts. In all instances there was a partial or complete obstruction of the common bile duct at its ampullary extremity by a biliary calculus. In several of the cases in which more than one cholangiographic examination was performed, the renal pelves were filled with the contrast medium at one examination but not at another, although the biliary ducts were well distended at the time of each cholangiographic observation. This apparent inconsistency is believed to be due to a failure to exert sufficient pressure during injection to cause regurgitation from the biliary radicles into the blood stream.

In the opinion of Rigler and Mixer the phenomenon of regurgitation occurs more frequently than their particular series of instances indicates. The renal areas are often obscured on roentgenograms by gaseous and other intestinal residues, particularly those that are opaque to roentgen rays, and small concentrations of opaque liquids in the renal pelvis are easily overlooked. They pointed out that optimal conditions must prevail if the phenomenon is to be observed.

Three possible explanations were offered for this rapid entry of the cholangiographic medium into the blood stream and its excretion by the kidney in a concentration sufficiently high to make the renal pelvis visible on roentgenograms. Absorption from the intestine was not regarded as a plausible explanation because obstruction at the lower end of the common bile duct was demonstrated in all cases, and in no instance was an appreciable amount of the medium observed in the intestine. Direct absorption of the medium through the mucous membrane of the biliary ducts was thought to be improbable for several reasons. In the first place, evidence that the medium had entered the blood stream at all was elicited in but a small percentage of the cases in which a well-distended biliary duct system was demonstrated by cholangiography. If the mucosa of the biliary ducts were capable of ab-

sorbing significant amounts of the medium then every cholangiogram revealing a well-distended set of biliary ducts should also reveal the medium in the renal pelvis. In the second place, the medium was observed to enter the kidneys very rapidly, in one case as early as 5 minutes after it was injected. The experimental work of Shafiroff and Bierman[11] was cited in support of the proposition that absorption of the medium by the mucous membrane of the biliary ducts takes place slowly, if at all. They ligated the cystic duct of cats and carefully preserved the blood vessels and lymphatics of the gallbladder. They then injected various radiopaque substances into the lumen of the gallbladder. Crystalloid substances such as sodium iodide and most of the organic compounds of iodine commonly used in excretory urography, of which diodrast is an example, were absorbed from the lumen of the gallbladder only after 6 hours, and they appeared in the kidneys hours later. Rigler and Mixer pointed out that this represents absorption by the mucous membrane of the gallbladder and they thought it reasonable to assume that absorption by the mucous membrane of the biliary ducts would be even more markedly delayed. For these reasons they held that, at best, an insignificant amount of the cholangiographic medium was absorbed by the mucous membrane of the biliary ducts, and they concluded that the mode of entry of the medium into the blood stream in quantities sufficient to be roentgenographically demonstrable in the kidneys was directly into the hepatic sinusoids and veins and thence into the blood stream. Apparently the pressure under which the cholangiographic medium was injected in the cases in which this regurgitation occurred was too great for the walls of the biliary canaliculi to tolerate. The speed with which the medium gained access to the kidneys and the amount of medium demonstrated there indicate that rupture of the canaliculi must have occurred. In his discussion, Rigler mentioned that the more familiar phenomenon of pyelovenous backflow observed not infrequently in retrograde pyelography is an analogous situation.

In addition to providing another and a very graphic link of evidence for the existence of a relatively easy retrograde communication betwen the biliary ducts and the blood stream, especially under conditions of elevated intraductal pressure, this observation of Rigler and Mixer offers a new and very plausible explanation for some of the unfavorable reactions which occasionally are associated with cholangiography. Allergic types of reactions like those observed sometimes, and still too frequently, when the organic compounds of iodine prepared especially for use in excretory urography are administered by the intravenous route, are observed rarely, if at all, after cholangiography. Rigler encountered none over a period of 6 years in which he used diodrast for this purpose, and he said that he was aware of no report of such a reaction in the literature. Rigler and Mixer said that mild or even severe febrile reactions were not uncommon after cholangiography in their experience. They intimated that a mild or transient bacteremia might be responsible for those reactions. Such reactions occur only in cases of infection of the biliary tract. They reasoned that if the cholangiographic medium can be regurgitated into the blood stream it is also possible to force the bacteria present in the biliary ducts into the blood stream. Experimental work on animals, unpublished as yet, which was done in their laboratory, supports this contention. They discredited the idea that simple distention of the biliary radicles might be a cause of these febrile reactions. In none of the cases in which they demonstrated regurgitation of the cho-

langiographic medium into the blood stream was there evidence of infection of the biliary tract, and in none of these cases was there evidence even of mild unfavorable reaction. In all of them however, the biliary ducts were well distended. Neither could they agree with authors who attribute the severe reactions to regurgitation of the medium and presumably other possibly infectious content of the biliary ducts into the pancreatic duct. Their collaborator, Bergh[2], found that the pancreatic duct received the contrast medium in about a third of a series of cholangiograms he studied. Since the incidence of unfavorable reactions after cholangiography is much lower than this, Rigler and Mixer contended that the entry of cholangiographic medium and, with it, other contents of the biliary ducts into the pancreatic duct is probably not responsible for the postcholangiographic difficulty.

Very appropriately, Rigler and Mixer issued the warning that great care should be exercised in performing the necessary maneuvers of cholangiography. Injection of the medium should be made at the lowest possible pressure to obtain moderate filling of the intrahepatic ducts. They suggested that the medium might best be injected under roentgenoscopic control to facilitate the use of the minimal amount of pressure and yet realize the full benefit of the diagnostic procedure. They also insisted on the use of all possible aseptic precautions in the preparation and administration of the cholangiographic medium.

2. *A roentgenologic cue to the diagnosis of spontaneous hypoparathyroidism.* Camp[4] recently reported the results of some investigations he made on the significance of symmetrical calcification of the cerebral basal ganglia. He found 12 cases in which this phenomenon occurred and in which there was definite clinical evidence of parathyroid insufficiency and tetany. In general, the patients' symptoms were those which commonly complicate hypoparathyroidism, cataracts, convulsions, mental retardation and trophic changes. Four of the patients were males; 8 were females. At the time of onset of symptoms, 7 of the patients were less than 10 years old, 3 were between 10 and 20 years of age, 1 was 39, 1 was 42. At the time of Camp's examination, however, 1 patient was 2½ years old, 4 were between 10 and 20 years of age, 3 were between their 20th and 30th years, 1 was 32 years of age, and 3 were more than 40 years old. In 11 of the cases the parathyroid insufficiency was spontaneous; the other patient underwent thyroidectomy at the age of 19 years and hypoparathyroidism followed subsequently.

The pathogenesis of the calcification is fairly well understood. Originally, a deposition of colloid takes place in and around the smaller cerebral arteries, especially in the media and adventitia. The veins are rarely affected. Later, the deposits of colloid become calcified and, as they coalesce, perivascular sheaths and concretions are formed. When the process is extensive the capillary vessels may be occluded, but the lumens of the arteries are rarely diminished. Changes like these are frequently observed at necropsy, especially in the globus pallidus and in the dentate nucleus of the cerebellum, and when they are of minor degree they are often thought to have little significance. The pathologic process described may become marked at any age, and in either sex and in response to many diseases, not all of which are associated with neurologic symptoms. Camp and those who worked with him on this project expressed the opinion that these changes have been most marked and, therefore, most subject to roentgenolo-

gic detection in cases of parathyroid deficiency and other diseases featured by mental deterioration, sometimes with and sometimes without convulsions or motor symptoms referable to the extrapyramidal system. They did not hold the calcification responsible for the mental deterioration and convulsive seizures, because adequate treatment of the parathyroid insufficiency improves the mental condition of the patients and stops the convulsions; but the cerebral and cerebellar calcification undergoes no roentgenologically demonstrable change.

The roentgenologic appearance of symmetrical calcification of the cerebral ganglia was described as a distinctive one, although the degree and extent of demonstrable calcification vary within wide limits corresponding presumably with the stage of the disease that is present at the time of roentgenologic examination. At the stage when the calcareous deposits are microscopically small, they are expected to escape detection on roentgenograms made during life. The earliest demonstrable deposits Camp observed were small, irregular and discrete, and they were distributed symmetrically in the regions of the several basal ganglia, especially in the putamen and the caudate nucleus. With more extensive calcification, especially when it occurred in the dentate nucleus of the cerebellum and in the folds of the cerebellar hemispheres, the shadows assumed a wavy linear appearance. With coalescence of calcareous masses the shadows were larger, denser and more obvious. Calcified deposits in the cerebellum and in the deeper layers of the cerebral cortex were observed to occur coincidentally. They sometimes were so marked as to simulate vascular lesions or calcified gliomas. Calcification limited to the basal ganglia of 1 side was observed in 2 cases and it was minimal in both cases. Neither patient had evidence of neurologic or parathyroid disease.

The bilateral and symmetrical arrangement of the deposits was the feature which served to prevent diagnostic confusion with cerebral neoplasm. Pneumographic examinations were occasionally used to clarify doubtful situations. Deposits of calcium in the choroid plexus were confusing only in anteroposterior projections of the skull; lateral projections of the skull, however, readily revealed the characteristic, more anterior position of calcareous deposits of the larger basal ganglia.

Camp was careful to point out that calcification of the cerebral basal ganglia is not found exclusively in cases of parathyroid insufficiency. In the course of his study he observed the calcification in patients who had had encephalitis, tuberous sclerosis, toxoplasmosis or mental deficiency since birth, and in some who did not have any clinical signs and symptoms referable to the condition. The observation, in a relatively short period, of 11 instances in which parathyroid insufficiency and calcification of the basal ganglia were associated was to him more than mere coincidence. Since parathyroid insufficiency responds so promptly and so well to treatment, the diagnosis should be established as frequently and as quickly as possible. Camp expressed the belief that at least an occasional mentally retarded or handicapped patient might be salvaged if all patients with known or suspected parathyroid insufficiency were subjected to roentgenologic examination of the skull, and if estimates of the concentration of serum calcium were made in all cases of symmetrical calcification of the cerebral basal ganglia.

3. *Some roentgenologic contributions to the diagnosis of vermiform appendi-*

citis. If a roentgenologist is asked to make a diagnosis of appendicitis, he makes use of diagnostic criteria in no way different from those used by other physicians. He takes a detailed history, makes a careful physical examination of the patient, and gives due consideration to special examinations of the urine and blood. Naturally, he is more "x-ray conscious" than many another physician would be, and if his roentgenologic equipment happens to be handy he will probably want to submit the patient to certain very simple roentgenologic examinations, none of which are rigorous enough or time-consuming enough to have an unfavorable effect on the patient's prognosis. The preliminary procedure would probably be a so-called scout film (preliminary survey) of the abdomen, preferably in anteroposterior and postero-anterior projections. His principal concern in this examination is the exclusion of certain abdominal abnormalities which produce clinical manifestations that simulate those of appendicitis. Urinary lithiasis usually, and biliary lithiasis frequently, can be disclosed by this simple maneuver; the existence, site, and usually the nature and significance of other intra-abdominal calcified masses can be determined; opportunity is given to establish the presence and to consider the significance of abnormal intra-intestinal and extra-intestinal accumulations of gas. If, by chance, the cecum contains enough gas to make this organ identifiable as such, the roentgenologist may wish to perform roentgenoscopy to correlate the abdominal tenderness and muscular reaction with the cecum, and by inference with the appendix.

Not many roentgenologists today place much reliance on the evidence of appendicitis elicited by administering opaque suspensions by mouth or by enema in the hope of making the lumen of the appendix visible on roentgenograms or on the roentgenoscopic screen. It is true that the exact intra-abdominal position of the vermiform appendix can be determined in this way; its mobility can be tested by manipulation and its anatomic relation to the cecum can be depicted. These determinations are possible, however, only if the appendix receives the contrast medium. This will be unlikely if the appendix is actually diseased in the clinical sense, although failure of the appendix to receive the contrast material is in no sense acceptable evidence of appendiceal disease. Nevertheless, to be able to demonstrate a precise correlation between significant abdominal tenderness and abnormal muscular reaction to palpation, and the tip of the cecum may, especially when the cecum is not in its usual position, be helpful in clarifying a dubious situation.

Even if there were distinctive roentgenologic signs of appendicitis, administration of opaque material by mouth would rarely be undertaken when the diagnostician is confronted with the possibility of acute appendicitis. Hours are consumed waiting for the material to reach the ileocecal region, and these are critical hours. Administration of opaque material by enema is much less time-consuming but it is not without danger when perforation is imminent, because it is a rigorous procedure, and no matter how deftly performed it excites the motility of the intestine at the very time when such excitement should be scrupulously avoided. In the interval between recurrent acute or subacute attacks of appendicitis, opaque material can probably be administered with relative safety by either route. But what is the diagnostic yield? Again it is true that lesions of the intestinal tract other than appendicitis, but perhaps simulating it in their clinical manifestations, will be detected in this way.

But aside from the apparently rare demonstration of an inspissated nonopaque fecal mass in the lumen of the appendix, or verifying the intraluminal or juxtacecal situation of an appendicolith impregnated with calcium, there are no reliable and distinctive signs of clinically significant appendicitis. The patient may complain of tenderness, when the cecum or the appendix is manipulated during roentgenoscopy and there may be some voluntary or involuntary abdominal muscular reaction on the part of the patient to the palpation, but at most these are signs of doubtful value and but a minor part of the clinical syndrome. If the appendix fills with barium suspension, and there is reason to think that 3 out of 4 appendixes will do this either with the barium meal or with the barium enema, or with both, the roentgenologic examiner may get the impression from his manipulations during roentgenoscopy, or even less reliably from a set of serial roentgenograms of the ileocecal region, that the appendix is fixed unnaturally to the cecum or to the abdominal parietes, and he may interpret this to be a residuum of previous attacks of appendicitis. Unfortunately the natural fixations of the appendix are most difficult to distinguish from the unnatural ones, in fact, many examiners of recognized competence experience difficulty in establishing merely the fact of fixation. Other more or less significant changes that may be observed in the barium-filled appendix are: (1) its infracecal, paracecal or retrocecal position; (2) diverticula, which are very rarely encountered and of questionable clinical and pathologic significance; (3) kinks and tortuosity, potential factors in the pathogenesis of acute appendicitis when it can be established that they are genuine, and this is not easy to do; (4) appendiceal stasis, the significance of which is impossible to estimate because it is known that apparently normal appendixes can retain roentgenologically demonstrable quantities of barium for days or weeks; (5) opaque foreign bodies, especially gunshot, which may be retained for prolonged periods; (6) small, nonopaque masses which may be small fecaliths or inspissated mucus, probably formed in situ, the significance of which is questionable; (7) peculiar and as yet unexplained changes in the relief pattern of the appendiceal mucous membrane.

Neoplasms of the vermiform appendix, malignant and benign, the incidence of which is exceedingly low, are occasionally made manifest not by producing demonstrable change in contour of the contrast-filled appendix, because the appendix is not expected to receive the contrast material when so affected, but by producing a compression or displacement type of deformity of the cecum or adjacent portion of aboral ileum. The tumefactive chronic periappendiceal abscess or granuloma produces a similar roentgenologically demonstrable change.

These remarks were prompted by the appearance in recent issues of radiologic journals[5 6 7 12] of several articles pertinent to the subject of appendicitis. Three of these called attention again to the significance of calcified appendicoliths. To those readers who accept the theory which Wangensteen[13] developed in a very convincing manner, namely, that obstruction of the appendiceal lumen is the fundamental factor in the pathogenesis of acute appendicitis and that the appendicolith is the most important single agency in the development of the obstruction, these discussions will have a special appeal. It is known, of course, that appendicoliths are frequently found in appendixes removed surgically for acute appendicitis (in 67% of a series of cases of acute appendicitis analyzed by Bow-

ers[3]), but the preoperative diagnosis has been made relatively seldom. The diagnosis is strictly a roentgenologic problem and it is a relatively simple one when the concretion is fairly large and when it is impregnated with enough calcium to make it opaque to the roentgen rays. The incidence of appendicoliths of this kind has been thought to be very low. Yet Felsen and Bernhard[7] encountered 10 instances in the course of 3 years during which time they estimated that 300 appendectomies were performed in the hospitals in which the 10 patients were seen. They also made an analysis of 100 cases reported in the literature since 1906 and of the 10 cases of their own. (See below.) Thomas[12] reported on the clinical histories and roentgenologic findings in 8 cases, 6 of which were observed since 1945 and 4 of them in that year. In Childe's[5] series of 8 cases the patients were children 12 years old or younger who were observed in a period of 11 years in 1 hospital. These reports are not mutually inclusive. In only 11 of the 100 cases reviewed by Felson and Bernhard[7] was the diagnosis made preoperatively and confirmed surgically. Summarizing the important conclusions which the authors of these papers independently concurred in making, these points can be made: (1) appendicoliths appear on roentgenograms of the abdominal field as oval or round or triangular or irregular opaque masses, which vary in size between 0.5 cm. in diameter to 3.0 by 1.5 cm. in length and width, and usually show evidence of lamination (94%, according to Felson and Bernhard), are situated in the far right lateral portion of the abdominal field at various vertical levels between the shadow of the lower border of the liver and that of the spine of the right ischium. (2) The shadows are large enough and opaque enough to be seen on the roentgenogram of the abdomen, although special projections are sometimes necessary to identify the shadows as appendicoliths and occasionally it is necessary to perform urographic and cholecystographic examinations to exclude urinary or biliary calculi or to examine the ileocecal region with the opaque meal or the opaque enema to prove a close anatomic relationship of the shadow with the cecum. (3) In about two thirds of the cases reported there was but 1 appendicolith; there were 2 or more appendicoliths in a third of the cases. (4) Not all appendicoliths are hard and firm; according to Felson and Bernhard about a fourth of them have a soft consistency. (5) Since the coincidence of acute appendicitis with appendicolithiasis is very high, and since the incidence and imminence of perforation of the appendix at or near the appendicolith is also very high, surgical removal of the appendix should be attempted as soon as the diagnosis is made; Thomas was especially insistent on this point and made the apt comment that it is more important to remove an appendix containing stones than to remove a gallbladder with stones since rupture of the gallbladder is a comparatively unusual complication of cholecystic disease. (6) Roentgenographic examination of the abdomen should be performed routinely whenever the diagnosis of appendicitis is being entertained.

Euphrat[6] reviewed the literature on mucocele of the appendix and reported a case in which, by using a well-defined roentgenologic syndrome elaborated by Akerlund[1], he was able to make the correct diagnosis prior to operation. The patient was a woman, aged 42 years, who for 10 months had had dull pain in the lower part of the right side of the abdomen while lying down. Physical examination revealed a smooth, round, slightly tender, very mobile mass at the level

of the anterior superior spine of the ilium. Roentgenologic examination of the colon showed the cecum to be displaced medially and anteriorly by a reniform, mobile, sharply circumscribed mass which measured about 6 by 10 cm. The mass maintained a close anatomic relationship to the cecum when manipulated, and it produced a concave deformity of the lateral and posterior borders of the ileocecal coil by its close contact. In the central portion of the mass there was an accumulation of calcareous material arranged in the shape of a swastika. No contrast material was observed to enter the lumen of the appendix. On the basis of these findings the diagnosis of mucocele of the appendix was made. The diagnosis was confirmed at operation. Qualitative tests of the contents of the mucocele were strongly positive for calcium but there was no trace of barium. This appears to be the fourth reported case of mucocele of the appendix in which the correct diagnosis was made before operation or necropsy. Euphrat expressed the opinion that in the past the diagnosis was usually missed because the presence of a mucocele was not considered. Undoubtedly there have been other instances, as yet unpublished, in which the diagnosis was made preoperatively and subsequently confirmed, for Akerlund's criteria are so definite and so readily elicited. The only feature of the syndrome distinctive for mucocele is the deposition of calcium in the substance of the tumor or in a plaque-like arrangement in its walls. Benign and malignant neoplasms of the appendix are manifested roentgenologically in a manner similar in all other respects. A well-localized chronic periappendiceal abscess may effect similar changes. Obviously the clinician and the roentgenologist should consider all the possibilities when alterations in the ileocecal region of this general nature are encountered in practice.

4. *Roentgenologic aid in the diagnosis of mesenteric thrombosis.* Some years ago, Rendich and Harrington[9] reported 3 cases of mesenteric thrombosis, in 2 of which they postulated a diagnosis of mesenteric thrombosis on the basis of observations made in the first case. In this case the patient was a woman, aged 75 years, who entered the hospital because of obstipation and abdominal pain of about 60 hours' duration. The clinical diagnosis was intestinal obstruction. Roentgenographic examination of the abdomen revealed distention of the small intestine and of the right half of the large intestine with gas. The column of gas in the large intestine ended abruptly at the transverse colon limb of the splenic flexure. This suggested mechanical obstruction; but, surprisingly, the opaque enema readily passed through the site of the suspected obstruction to fill the entire colon. The patient died the next day. Necropsy revealed thrombosis of the superior mesenteric artery; gangrene of the small intestine, cecum and part of the ascending colon, and hypertensive heart disease. In the course of the next nine months they observed two other patients who presented a similar intestinal distention on roentgenographic examination of the abdomen. On this basis alone they again offered the tentative diagnosis of mesenteric vascular occlusions. Both patients underwent operation. The diagnosis was verified, the gangrenous intestine was resected, and the patients recovered.

Recently Harrington[8] reported 4 additional cases with precisely similar roentgenographic manifestations in which the diagnosis of mesenteric thrombosis was confirmed either at operation or at necropsy.

The authors[8 9] did not say how often this diagnosis was made but disproved by subsequent events; therefore, they presumably did not have this experience. In any event, their published roentgenograms are convincing enough to make the clinician and roentgenologist give this diagnostic possibility due consideration whenever gaseous distention of the intestine abruptly terminating in the region of the splenic flexure of the colon has occurred. It is the region which corresponds to the limit of the distribution of the superior mesenteric vessels. Obviously, it is another situation in which a simple roentgenologic procedure may offer an important clue to an obscure and relatively rarely encountered cause of genuine abdominal catastrophe, and one for which prompt decision might spell the difference between life and death.

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PHARMACODYNAMIC CONSIDERATIONS OF ATROPINE AND RELATED COMPOUNDS

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Few drugs have enjoyed such wide or varied use as atropine and its related compounds. Since some ancient time, when plants of the belladonna group were first ingested, up to the recent flood of synthetic modifications it has been a basic tool of witch doctor, physician and scientist. What basis does modern pharmacology give us for separating fact from myth—for replacing empirical hearsay with applied physiologic observations? Partial answers to these questions are given in the following review—an outgrowth of some investigations on methyl atropine nitrate at the University of Rochester.

Chemical Structure and Biologic Activity. What is known of the relationship between the chemical structure and biologic activities of these drugs? The active principle of the belladonna leaf and its extracts are a group of alkaloids, the most important of which is l-hyoscyamine. Atropine is a stereo-isomer of hyoscyamine and is optically racemic. The compound is the ester of tropine (an amino-alcohol) and tropic acid (alpha-phenyl, beta-hydroxy-propionic acid). Extensive investigations and modification of both portions of this molecule have been carried out to determine if possible which chemical and structural features are essential for various biologic activities displayed by the parent alkaloid. Cushny[18] (1920) felt that tropine itself was devoid of "typical" atropine action. However, its esters (tropeines) with a great variety of acids possess some degree of biologic activity. This was systematically studied by Dyson[22] (1925). In general, he found that mydriatic activity was stronger in those compounds containing a benzene ring and/or an alcoholic hydroxyl group in the acidic portion of a molecule, but this was not the essential prerequisite it was often thought to be. For example alpha-hydroxy-beta-2 and pyridyl tropeine, which has no benzene ring portion, and ortho-hydroxy-benzoyl tropeine and meta - hydroxy - benzoyl tropeine which have no alcoholic hydroxyl groups, all possess mydriatic activity. Indeed the lactone itself possesses mydriatic activity as shown in terebyl tropeine which has paralytic activity in addition; thus this property does not depend on a free hydroxyl group either. Opening the lactone ring destroys activity.

The irritant action of atropine is diminished with the substitution of halogen (chlorine or bromide) for the hydroxyl portion of the atropic acid residue.

Alkyl atropines are without activity.

Demethylation of the nitrogen atom results in decreased activity.

Methyl atropines have little or no central action but retain their peripheral activity.

It has been shown, then, that certain alterations in structure modify various types of atropine actions. Thus syntropan (the dimethyl-amino-propanol ester of tropic acid) is quite effective in reducing spasm of smooth muscle (though 20 times less effective than atropine itself in equivalent doses) it is almost without effect upon the central nervous system; it does not suppress pilocarpine, induces salivation, and is a very weak mydriatic.

Novatropine (methyl-homatropine hydrobromide) was shown by Quigley[79] (1937) to be somewhat less effective than atropine in inhibiting gastric hypermotility induced by insulin.

Homatropine is an excellent mydriatic, although somewhat transient in effect but has little systemic effectiveness, possibly because the body esterases are able to destroy it rapidly (Bernheim,[6] 1942).

Eucatropine (4-mandelyloxy-1; 2; 2; 6-tetramethyl piperdine) has also been used clinically as a mydriatic.

Mydriasin is a trade name for methyl-atropine bromide.

Methyl-atropine nitrate is sold in this country under the trade names of eumydrine and metropine.

A long series of benzilic esters were investigated by Ing *et al.*[50] (1945) and they briefly reviewed the problem. Their observations covered compounds of the following general type:

$$\text{Acyl} - \text{O} - (\text{C})_n \pm \text{N}\begin{cases}\text{R}^1\\\text{R}^2\\\text{R}^3\end{cases} \overline{\text{X}}$$

which structurally are all alkamine esters where the $(C)_n$ group is either a polymethylene chain or a cyclic structure. It is a striking fact that compounds of this general type have a peculiar affinity for neural structures and exhibit 1 or more of 4 kinds of pharmacologic actions, namely, cholinergic, atropine-like, curare-like and local anesthetic, depending on the nature of the acyl radical and of the basic group. The discussion of these writers is worth quoting: "Cholinergic properties appear to be rather strictly confined to molecules containing 3 methyl groups on the N-atom and an aliphatic acyl radical (acetyl, pyruvyl, carbamyl, etc.); the $(C)_n$ group is usually $(CH_2)_2$ but n may be 3 or the chain branched, *e. g.*, acetyl-methylcholine."

"Atropine and curare both antagonize acetylcholine drugs but the anatomic distribution of their effects is different. Curare-like properties are widely shared by alkamine esters, provided that the basic group is quaternary, but they may appear in a milder and more transient form in tertiary bases. The curare type of activity is characteristic of the onium cation and when it is displayed by tertiary bases most probably depends upon a relatively stable tertiary cation $R_3\overset{+}{N}H$, because strong bases show it more markedly than weak bases.

"The results, which are reported in this paper, suggest that atropine-like properties ought also to be attributed to the cation and not to the free base. Conversion of a tertiary base into its metho-salt stablizes the cation. Whereas tertiary cations $R_3\overset{+}{N}H$ exist in equilibrium with the free base, thus: $R_3\overset{+}{N}H + \overset{-}{O}H = R_3N + H_2O$, quaternary cations are incapable of such a reversible reaction. We have found that such stabilization normally increases the atropine-like properties of tertiary bases. The attribution of atropine-like properties to the cation of the tertiary base not only explains our results but also brings the atropine-like action into line with the curare-like action; both are due to antagonism of an onium cation, acetylcholine, in cells adapted to respond to the latter, and our view is that the cationic nature of the antagonist, whether atropine-like or curare-like, is fundamen-

tally important. The conversion of a tertiary base with atropine-like properties into its metho-salt not only increases its activity in this respect, but also confers upon it strong curare-like properties, as Brown and Frazer[10] (1869) discovered for atropine itself. The other structural features which lead to atropine-like properties in alkamine esters are more restricted than those which confer curare-like properties."

Physiologic Activity of Atropine. In the previous section, while considering chemical structure, the term "typical" atropine activity was glibly used. Just what is typical atropine activity?

The most prominent physiologic action of atropine may be expressed as antivagal, that is, it tends to block the effects of acetyl choline on smooth muscle and gland cells, leaving adrenergic action dominant and unopposed. That atropine often selectively blocks the muscarinic action of acetyl choline without affecting the nicotinic effects of acetyl choline is illustrated by Frey's[29] experiment (1928) on the intestinal tract of the tench. Clark[12a] (1937), however, offers well documented evidence to show that this is a generalization, and every tissue seems to be a law unto itself. For example, in *Helix pomatia* the heart action is inhibited by acetylcholine or muscarine. The presence or absence of atropine has no effect on this action. One becomes accustomed to thinking of atropine as a parasympatheticolytic drug with solely peripheral action, but these drugs have definite central nervous system and sympathetic components in their action. It is worth recognizing that although little is known about the exact way in which atropine works, there is abundant evidence that in no instance does it prevent nerve impulses from releasing acetylcholine but merely prevents effector cell response at some point peripheral to the nerve ending.

Action of the Atropine Group on the Central Nervous System. Although the autonomic nervous system is customarily thought of in terms of peripheral effects there is much evidence to show that the concept of levels of function applies to this system as well as to the motor division. In isolated organs effects can be produced by local application of specific autonomic drugs. Similar effects can be produced in intact animals, but segmental autonomic reflexes are well known and medullary centers are essential for regulation of the blood pressure. Heat regulation, primarily an autonomic function, is integrated at the level of the hypothalamus. Indeed, injection of acetylcholine (pituitrin, or pilocarpin; adrenalin produces no effect) directly into hypothalamus results in massive parasympathetic discharges which can be blocked immediately by intraventricular injections of atropine (Henderson and Wilson,[47] 1936; Cushing,[16] 1932). Other more complex integrations take place at the striatal level. Cortical areas governing autonomic function are found, generally corresponding in localization to those areas influencing corresponding somatic motor functions. For example, lacrimation occurs from stimulating eye-fields; salivation on stimulating motor representation of the face and tongue. The exact anatomic or chemical pathways through which these effects are mediated are not known. Heart rate, intestinal motility, sweating, conditional reflex salivation and psychic phase of gastric secretion are all known to be influenced by autonomic stimuli and also by cortical stimulation. The relationship that acetylcholine, and consequently atropine, might have in these central nervous system functions is not well understood at the present time.

Atropine stimulates the medulla and higher cerebral centers. It is believed that the drug acts by direct irritant action and not by removal of inhibition. Many people believe that this action on the central nervous system is the basis of the extreme toxicity of atropine in human beings as compared with other animals, notably rodents. Macht[63] (1923) studied

the effect of different doses of atropine on the reactions of albino rats in a circular maze. He found that very small doses would affect ability in this situation, 0.025 mg. stimulating and 0.05 mg. depressing their activity. The depressant dose of tropine was 5 mg., while tropic acid had no effect even in large doses. These results were obtained with rats: animals in which the lethal dose of atropine is relatively high and in which central nervous system effects are not prominent. On the other hand, psychic symptoms are among the most prominent signs of atropine intoxication in man leading to many grotesque and unusual actions. The intra-arterially injected acetylcholine causes a discharge from the central nervous system of a decerebrate cat resulting in an increase in tone of the innervated quadriceps. This action is blocked by atropine (Calma and Wright,[32] 1944).

Methylatropine possibly by virtue of its methylated structure (Dyson,[23] 1928) has equivalent peripheral action but little or none of atropine's prominent central action. Mice are killed by respiratory paralysis without any evidence of convulsant action (Graham and Lazarus,[38] 1940). Nyman reports that Brown and Frazer[10] (1869) found that the stimulating effect of methylatropine (iodides and sulfates) had considerably less central action than equivalent doses of atropine. Erbe[27] (1903), in extensive clinical trials, failed to find evidences of central stimulation with therapeutic doses in man equivalent to those which had an action, when tested in a cat's eye, as great as that of atropine. Issekutz[51] (1917) found that methylnitrates of both homatropine and atropine has stronger anticholinergic effects than the parent drugs and showed a marked curare effect but that the central effect was 30 to 50 times weaker. Solis-Cohen and Githens[83] (1928) state that ingestion of 60 mg. of methylatropine nitrate by a man caused weakness, particularly of the legs, but no cerebral symptoms. Monrad[71] (1928) reports, however, nystagmus, restlessness and convulsions as among the toxic reactions to methyl-atropine nitrate in his series.

Various belladonna derivatives have been used clinically for many years in paralysis agitans, athetosis and rigidity due to involvement of the extrapyramidal system and the basal ganglia. In general, little is understood as to their mechanism of action. Atropine is used for Parkinsonian symptoms, but the prominent side effects are often produced before alleviation. Highest recorded daily dose of atropine in this condition is 54 mg. (Hall,[40] 1937). No published results concerning methylatropine therapy in Parkinsonism were found.

Neither atropine nor methylatropine shows any regular action on the spinal cord.

Therapeutically this means that methylatropine has less prominent psychic side actions, while maintaining a peripheral effect in equivalent doses.

Action of the Atropine Group on the Eye. Atropine causes paralysis of the sphincter muscle of the iris which dilates the pupil (mydriasis). It also paralyzes the ciliary muscle of the lens leading to paralysis of accommodation (cycloplegia) and photophobia. The iris no longer responds to light and fails to react with convergence of the eyes. The parasympathetic fibers from the tectal outflow reach the eye *via* the third nerve and the ciliary ganglion and innervate the constrictor pupillæ and the ciliary muscle, and form the efferent limbs of the light and accommodation reflexes. This reaction takes place by blocking the cholinergic nerve impulses and the action of acetylcholine. Intraocular pressure remains essentially unchanged, unless it is already elevated as in glaucoma, when it is increased further with atropine. There is some debate as to the mode of action of atropine in increasing ocular pressure in such circumstances. It is probable that lymphatic drainage is still more seriously interfered with by the action of atropine

on the ciliary muscle, blocking free flow between the 2 chambers of the eye.

Methylatropine was introduced clinically for mydriatic use by Lindenmeyer[59] (1903) and Goldberg[36] (1903). Their studies indicated that methylatropine produced maximal mydriasis in 30 to 75 minutes with final recovery in 40 to 72 hours. It produced maximal cycloplegia in 2 to 14 hours with recovery in 37 hours. These results may be compared with the recent results of Wolf and Hodge[103] (1946), who tested the effect of methylatropine and comparable solutions of homatropine and atropine in the human eye. Instilled in equal amounts into human eyes, homatropine had less mydriatic and cycloplegic effect than either of the other 2 drugs, and recovery from its effects was the most rapid.

Methylatropine nitrate had significantly less mydriatic action than atropine. The cycloplegic action was about the same as that of atropine.

Ing, Dawes and Wajada[50] (1945) report that mydriatic activity of methylatropine was 2.88 times as strong as atropine when given intraperitoneally to mice but only half to equally as active with local application to the cat's eye. They felt that activity of these alkaloids depends on the method of administration.

Action of the Atropine Group on the Respiratory Tract. Atropine inhibits secretions of the nose, pharynx and bronchi, with resultant drying of mucous membrane. According to Henderson[45] (1923) 0.2 mg. causes decrease in nasal secretion without drying of the mouth in a 65 kg. man.

Elftman[25] (1943) describes autonomic innervation of bronchial smooth muscle. Parasympathetic fibers are vagal, reach the bronchial tree from anterior and posterior pulmonary plexi and exert a tonic constrictor action; vagal section is followed by bronchodilatation. Stimulation of the peripheral end of a cut vagus produces ipsilateral and contralateral constriction. In animals, constriction *via* vagus may be reflexly invoked by irritating the mucous membrane of the upper respiratory tract.

Atropine also inhibits the smooth muscle of the bronchi and bronchioles, causing relaxation (Trendelenburg,[88] 1912; Sollmann and Gilbert,[84a] 1937). These actions form the basis of a therapeutic use in asthma and catarrhal conditions and of its use preoperatively with morphine when anesthesia is given.

No specific references to the actions of methylatropine on the respiratory system were discovered, but on the basis of the similarity of its other peripheral actions to those of atropine, it probably has a similar effect on the respiratory system. Usener[92] (1926) used dosages of 0.5 to 0.75 mg. of methylatropine nitrate for asthmatics.

Action of the Atropine Group on the Heart. Ever since the Webers[97] (1845) showed the inhibitory action of the vagus on the heart, there has been physiologic debate as to the exact rôle that the vagus nerve plays in controlling the activities of the heart. Hence there has been a similar confusion over the exact rôle of atropine; the essential action on the heart is antivagal, despite some claims for direct action of heart muscle. It is now generally believed that atropine alters the cardiac rate by stimulating medullary vagal centers in small doses of between 0.5 and 1 mg. (Heinekamp,[44] 1922; Gremels,[39] 1936). Recently accelerator fibers have been described in the vagus nerve of a dog (Kabat,[55] 1939) which may explain the initial slowing action of atropine. Atropine causes acceleration by blocking the vagal effect on the sino-auricular pacemaker in doses over 2 mg. (Crawford,[14] 1923; Marris,[67] 1916). These effects are most prominent in young adults who have a strong vagal element. White[98] (1940) reports that the bradycardia resulting from stimulating the hypothalamus in humans was abolished by atropine. He also obtained acceleration from stimulating the posterior portion of the hypo-

thalamus. Atropine is sometimes used in Adams-Stokes syndrome to prevent vagal arrest. It will prevent the bradycardia caused by giving acetylcholine. Atropine is occasionally effective in heart block (Gilchrist,[35] 1933).

Issekutz[51] (1917) found that methylatropine acts about 8 times as strongly as atropine in depressing the vagus of the frog. Nyman's[75] (1942) results (Tables 1, 2 and 3) working with atropine and other anticholinergic substances, demonstrate clearly that their effect on blood pressure, pulse rate and minute volume of the heart is most prominent in individuals with a strong vagal component. Atropine counteracts the peripheral vasodilatation and drop in blood pressure caused by acetylcholine and mecholyl. In toxic concentrations, however, it causes a drop in blood pressure because of central vasomotor depression. Despite its general effect of counteracting the vasodilatation, as previously mentioned, in certain cutaneous blush areas it causes vasodilatation perhaps by direct vascular effect. Methylatropine has been shown to influence the action exerted by adrenalin on the blood-vessels and has the power to cause paresis and ultimately complete paralysis of sympathetic motor termination in the vessels (Bergengren,[4] 1925). Graham and Lazarus[38] (1940) found that in rabbits 0.05 mg. per kg. of methylatropine nitrate inhibited the bowel without affecting blood-pressure; however, 6 μg. of atropine

TABLE 1.—THE EFFECT OF ATROPINE AND EUMYDRIN ON PULSE RATE

Subject	Drug	Dose (mg.)	Dose μ gn. 10 kg.=	Resting pulse	Maximum increase (%)
N, male	Atropine	0 84	100	50	20
	Methylatropine	0 84	100	52	73
O, male	Atropine	0 72	100	58	28
	Methylatropine	0 72	100	55	73
H, female	Atropine	0 65	100	85	21
	Methylatropine	0 65	100	84	46
P, female	Atropine	1 20	200	43	114
	Methylatropine	0 60	100	43	175

TABLE 2.—THE EFFECT OF ATROPINE AND EUMYDRIN ON THE OXYGEN CONSUMPTION, BLOOD PRESSURE, PULSE RATE AND MINUTE VOLUME OF THE HEART IN HUMAN BEINGS

Substance	Dose (mg. per kg.)	O_2 per ml. per min.	Pulse rate	Blood pressure	Min. vol. of heart (liters)
Atropine	0 02				
(before injection)	..	239	54	129/90	3 4
30–40 min.	..	242	84	136/96	4 0
60–70 min.	..	246	78	140/98	maximum increase (18%)
Eumydrin	0 02				
(before injection)	..	236	54	122/83	3 2
30–40 min.	..	236	85	137/92	3 7
60–70 min.	..	222	98	143/93	5 7 maximum increase (78%)

TABLE 3.—CHANGES IN SKIN TEMPERATURE RECORDED WITH THERMOCOUPLE AFTER EUMYDRIN

Location	Temperature before injection		Temperature after injection	
	30 min.	20 min.	20 min.	30 min.
	(0.1 mg. eumydrin/10 kg. body weight)			
Forehead	34 8°C	35 1°C	36 3°C	36 3°C
Right cheek . . .	34 2	34 3	36 8	36 0
Left cheek	33 9	34 0	36 7	35 8

0.2 mg. atropine caused responses of approximately the same magnitude when measured in the arms and hands.

abolished the effect on the blood-pressure of 0.15 μg. of acetylcholine for an average 9 minutes. Six μg. of methylatropine nitrate abolished the effect of 0.15 μg. of acetylcholine for over 19 minutes. They were impressed with the more lasting effect of methylatropine in depressing cardiovascular autonomic cholinergic fibers.

It will be seen here, likewise in man, that methylatropine nitrate has a more marked effect on the blood-pressure and minute volume of the heart than equivalent doses of atropine.

Action of the Atropine Group on the Salivary Glands. It has long been known that the atropine alkaloids decrease salivary secretion. This antisialogogic action is made use of in routine preoperative medication. Hedenheim[42] (1872) demonstrated that stimulation of chorda tympani after atropine no longer produced secretion of saliva from the sublingual gland. Langley[56] (1890) demonstrated that stimulation of the nerves leading to the parotid gland also did not produce salivation after atropine. Ewing[28] (1911) demonstrated that atropine decreased the quantity and composition of saliva. In greater doses, first the solids and then the amylolytic power of the saliva were reduced and that in the reverse direction normal amylolytic power returned first. It has been adequately demonstrated that whatever controls the secretory function, the effects produced are independent of those related to blood flow through the gland. The ability of these alkaloids to inhibit salivary secretion induced by pilocarpine has long been used as a method of testing the relative potencies. Issekutz[51] (1917) who used 20 mg. of pilocarpine in rabbits found that 0.5 mg. of methylatropine was as effective as 1.5 to 2.0 mg. of atropine, thus making the quaternary base 3 to 4 times stronger than the parent alkaloid. Cushny[18] (1920) found that methylatropine (bromide) was $1\frac{1}{2}$ times as strong as atropine in antagonizing pilocarpine induced salivation in dogs. Drugs were calculated as base. The dogs had fistulas in the submaxillary ducts, a standard technique of Cushny's, fully described in the literature. He used methylatropine bromide with a melting-point of 222° C. One-tenth mg. of atropine sulfate was weaker than 0.1 mg. of methylatropine but stronger than 0.05 mg.

Bulbring and Dawes[11] (1945) found that methylatropine was more effective than atropine in antagonizing pilocarpine-induced secretion in dogs. Giving atropine an arbitrary value of 100, the lowest value obtained for methylatropine nitrate was 133, the highest was 250, the mean 190±21. Nyman, in his extensive review of the atropine group, tested the effect of various drugs on pilocarpine-induced secretion in man. He found that in 7 consecutive control experiments on a male subject, O, an average secretion of 28.6±0.45 gm. of saliva in the 1st hour followed injection of 0.1 mg. of pilocarpine per 10 kg. body weight. Similar results were obtained in other individuals: 23.15 gm.; 24.8±0.57 gm.; 33.7±0.81 gm.; 30.65 gm. If subject O was given 0.05 mg. methylatropine nitrate (7 mg. per 10 kg. body weight) secretion was reduced to 14.95 and 15.55, in 2 different trials.

The following amounts of atropine sulfate antagonized pilocarpine-induced secretion (1 mg. pilocarpine per 10 kg. body weight). In subject O, whose unantagonized average secretion (given above) was 28.6 with 0.1 mg. (14 μg.) the secretion was reduced to 14.35. Dose of 0.2 (28 μg.) allowed secretion of only 9.7. Dose of 0.45 (63 μg.) allowed secretions of 1.95 and 1.3 gm. in 2 different trials. Dose of 0.5 (70 μg.) allowed secretion of 0.55. A dose of 0.6 mg. (84 μg.) completely stopped secretion. Results with methylatropine nitrate in the same individual were as follows: dose of 0.07 (10 μg.) allowed secretion of 12.15; dose of 0.1 (17 μg.) reduced secretion to 5.25 (35 μg.) inhibited secretion completely.

Note that this dose is only one-third the dose of atropine required to obtain the same antisialogogue effect. Similar results were obtained with 4 other individuals. Nyman charted and compared the activity of 14 other alkaloids with that of atropine in this regard.

Action of the Atropine Group on the Esophagus. Little information was discovered relating to the pharmacologic basis of the use of the atropine series in cases of cardiospasm. However, as early as 1881 Szpilman and Luchsinger[86] showed that the spasm produced by vagal stimulation resulting in contraction of smooth muscle in the lower third of the esophagus was depressed by atropine. The striated muscle of the esophagus apparently is not under autonomic regulation and, therefore, not affected by atropine. With the recent establishment of vagotomy as a frequent operation in duodenal ulcer patients, it is probable that more information relative to vagal control of the esophagus in man will be obtained.

Others believe that cardiospasm, pylorospasm and megacolon are results of sympathetic not parasympathetic denervation and arise from the inability to relax rather than from spasm of the sphincters. It must be reiterated, however, that response of the entire gastro-intestinal tract to autonomic stimulants depends on the physiologic state of the organs and the organism, and that general conclusions as to the reactions are dangerous.

Action of the Atropine Group on the Stomach. The work of Bastedo[3] (1936) and his article on the value of atropine and belladonna in stomach disorders with its accompanying references summarize this problem as of 1936. Here, too, much additional information will be gained on the vagal effects controlling the stomach from vagotomies. It has been thought since 1896 with the work of Ushakoff[91] that very large doses of atropine in animals will lessen gastric secretion and lower the acidity. After vagus and splanchnic nerves have been severed, the stomach continues to function, frequently with little if any dilatation; therefore its tone and rhythmic motility must be intrinsic, and the extrinsic nerves may be only regulatory, thus giving a more delicate control. The vagi and splanchnic nerves are for the most part not motor antagonists. The general effect of vagal stimulation is tonic while that of splanchnic stimulation is tonic to the pylorus and relaxing to the stomach wall. But stimulation of either may result in increased tone when the stomach is hypotonic, and in decreased tone when the stomach is hypertonic. Because these nerves are not a balanced mechanism in control of the gastrointestinal tract, abolition of one does not necessarily result in overactivity of the antagonist. Atropine may act on all nervous structures but its most regular action is to render vagal stimuli ineffective.

The effect of atropine on gastric secretion is best considered in relationship to the various phases of activity. Secretion occurring in the psychic phase was abolished by atropine in animals. Doses of $\frac{1}{50}$ grain reduce the amount of secretion in the psychic phase but does not abolish it in humans. It is quite possible that this physiologic atropine reaction is the basis of decreased appetite in humans following atropine. There is some evidence that the intestinal phase of gastric secretion is partly mediated through the vagus in animals and that this effect may be abolished by atropine (Ivy,[52] 1935). The effect of atropine on the chemical phase of gastric secretion has been the subject of much debate. Bastedo[3] believes that there is some reduction in free and total acid with section of the vagi, and with doses of atropine the equivalent of 2 grains for humans, cats show a decrease in secretion as well as in acid and mucin content, but the secretion is not completely abolished. Generally, patients tend to show a decrease in the volume of

secretion following atropine, without a corresponding decrease in acid. After a dose of 2 mg. of atropine sulfate (sufficient to produce dryness of the mouth, blurred vision, tachycardia and cerebral symptoms), the total volume was reduced, but a test breakfast of alcohol produced a normal amount of secretion. Large doses of atropine depressed the mucin content before affecting the acid content, a dangerous effect from a standpoint of ulcer patients in whom mucin has been shown to protect against irritation by acid.

In summary, the enormous doses of atropine possible in experimental animals may reduce and even abolish the secretion of gastric juice. In man the maximal therapeutic dose tends to reduce the psychic phase and possibly the intestinal phase of gastric secretion and thereby reduces the total secretion. It also tends to reduce the continuous interdigestive secretion for periods of 2 to 3 hours. In the chemical phase, doses large enough to cause toxic reaction may bring about a distinct reduction in the amount of secretion, though this is by no means a constant effect, and when present is short lived. If atropine is to be used in this manner, the minimal dose having any effect on the stomach is 1 mg. subcutaneously. Bastedo considers any smaller doses merely homeopathic.

Nyman studied this problem and was unable to show that methylatropine was any more effective than atropine in this regard.

The Effect of the Atropine Group on the Pylorus. Schroeder[81] (1933) believes that the normal innervation of the pyloric sphincter is tonic, rhythmic relaxation following the beginning of the antrum contraction. Stimulation by adrenalin inhibits antrum peristalsis and the rhythmic activity of the sphincter, but increases the rate of flow. Pilocarpine has the reverse effect. Atropine acts like adrenalin. Ephedrine as well as the vasopressor and oxytocic principles of the posterior pituitary lobe inhibit contraction. Morphine causes increased antrum peristalsis and contraction of the sphincter. Papaverine is more effective than atropine or posterior pituitary in antagonizing this morphine effect. Bastedo[3] (1936) believes that pylorospasm may be either splanchnic-vagal or psychic in origin, and that atropine is only effective in those due to increased vagal tone.

Treatment of Hypertrophic Pyloric Stenosis. These properties have been employed in the treatment of pylorospasm and infantile pyloric stenosis. White[101] (1923) developed the concept that some of these infants have a fundamental autonomic unbalance which atropine and methylatropine may be used to correct. Others speak of these infants as being hypertonic.

Effects of the Atropine Group on the Small Intestine. Autonomic control of the small intestine is essentially regulatory, and the intrinsic nerve networks are of much greater importance in the usual physiologic state. Consequently there has been much confusion in the literature of the exact rôle played by atropine. Henderson and Roepke[46] (1937) feel that much of this arises from the undue weight given to the work of Magnus[66] (1908), who claimed that atropine stimulated the bowel. Examination of his experimental techniques revealed that his results are based on experiments with intestinal strips using concentrations of 1:4000 in a medium which we now would regard as highly unphysiologic. They point to the work of Unger[89] (1907) who showed that atropine produced a decrease in tone and movements in a concentration of 1:2,000,000 and less. Comparison with doses long believed effective in man (1 to 2 mg.) demonstrates that these doses probably lead to concentrations of 1:70,000,000 if the atropine is distributed throughout the whole 60 kg. of body weight, or 1:5,000,000 if the atropine is distributed only in the blood. Toxic

doses for man range around 10 mg. Even here the concentration will be in the range of 1:1,500,000. Thus the use of a 1:4000 solution seems fantastic when attempting to describe the effects of therapeutic doses.

In human beings an excellent study by Elsom and Drossner[26] (1939) tended to confirm the opinions long held by clinicians. In the normal duodenum, intestinal tone is high and motor activity great. There are 2 types of waves: (*a*) large peristaltic waves occurring every 1 to 2 minutes and lasting approximately 1 minute; (*b*) superimposed on these are frequent small undulations produced by rhythmical contraction. Administration of $\frac{1}{150}$ grain of atropine was followed in 10 to 15 minutes by a gradual fall in intraduodenal tone. The large peristaltic contractions were altered simultaneously, often becoming more pronounced as the tone decreased, then gradually diminishing in size and frequency and eventually disappearing. The small rhythmical waves decreased in size, though not in frequency. The effect was fully developed at the end of 20 minutes and lasted for 1 to 2 hours, after which there was gradual recovery.

The normal action of the jejunum and ileum consisted of small rhythmic undulations which were definitely decreased in tone and character following atropine. These results were obtained by means of a balloon inserted within the lumen of the gut of several human subjects. These results agree with those reported by Herrin[48] (1936) and White[101] (1923) who maintain that gastro-enterospasm was a manifestation of autonomic unbalance in early infancy and was corrected by administration of atropine.

There have been no experimental studies of the response of the gut in human beings using methylatropine other than those discussed in connection with pyloric stenosis. However, Handovski[41] (1929) tested the effect of methylatropine nitrate, atropine and papaverine on isolated strips of mouse intestine. He found that approximately 2 cc. of 0.2% solution of methylatropine hydrochloride would inhibit contraction, whereas approximately 6 cc. of 0.33% solution of atropine sulfate was required to accomplish the same effect; $2\frac{1}{2}$ cc. of papaverine hydrochloride (0.33% solution) would also inhibit contraction. However, papaverine and methyl-atropine nitrate seem to have synergistic actions, so that only 0.8 cc. of an equal mixture of 0.43% papaverine hydrochloride and 0.007% solution of methyl atropine hydrochloride were necessary to inhibit contraction. Graham and Lazarus[38] (1940) found that the spasmolytic powers of atropine and methyl-atropine nitrate were comparable; both drugs produced reduction in tone and loss of segmental movements of the intestine of rabbits anesthetized with ether. Both drugs antagonize the neurogenic spasm inducted by eserine. The minimal effective concentration of both for rabbit jejunum was 1 part in 10,000,000. There is a definite inhibition for 2 or 3 minutes with dilutions of 1:5000,000. Both drugs were without effect on spasm produced by barium chloride which is a myogenic stimulant. The range of sensitivity of rabbit intestine to these drugs was the same and depressed upper jejunum, duodenum, ileum and colon in that order.

The Effect of the Atropine Group on the Colon. Doses of atropine between 0.4 and 0.6 mg. decreased the tone and activity of the colon for $1\frac{1}{2}$ to 2 hours, with gradual return to normal (Elsom and Drossner,[26] 1939). Jackmann and Bargen[54] (1938) stated that doses of 0.0006 gm. produced definite depressing effects on colonic activity but had marked side effects which made it undesirable for routine clinical use.

Schroeder[81] (1933) studied the effects of various drugs on the ileocecal valve and ileum. He believed that normally the sphincter undergoes tonic rhythmic relaxation following the beginning of ileal

peristalsis. Adrenalin inhibits peristalsis of the ileum and rhythmic activity of the valve but increases the rate of flow. Pilocarpine has the reverse effect. Atropine acts in the same manner as adrenalin.

OTHER EFFECTS OF THE ATROPINE GROUP ON SMOOTH MUSCLE. *Ureter.* Atropine has an antispasmodic action on the ureter when it is in spasm and will decrease the increased tone resulting from morphine in man (Macht,[62] 1918). No reports were found dealing with the effect of methylatropine on the ureter.

Biliary Duct and Gall Bladder. The atropine group exerts a mild antispasmodic action and strong enough to counteract contractions induced by morphine. Both nitrites and xanthins were effective against morphine contractions (Lieb and McWhorten,[57] 1915; Macht,[61] 1917).

Uterus. Actions of the atropine group are of little demonstrable effect on uterine contractions, despite its wide use clinically in dysmenorrhea (Novak,[74] 1915). They are, however, able to counteract such effects as are produced by acetylcholine and its derivatives.

Urinary Bladder. The atropine group has a depressive effect on the detruseus muscle, especially when it is hypertonic (Edmunds and Roth,[23a] 1920). MacCallum[60a] (1906) reported that atropine had little effect on urine flow in rabbits but antagonized the depressant effect of pilocarpine. (See also Featherstone, R. M., and White, N. G., *J. Pharm. and Exp. Ther.*, 84, 105, 1945.)

The Effect on Sweat Glands. What long seemed a paradoxical action of the atropine group in inhibiting sweating (glands known to be supplied by sympathetic nerves) was cleared up with the discovery by Dale and Feldberg that acetylcholine was released by these nerve-endings, notwithstanding their anatomic relationships. Howe studied the effect of atropine in inhibiting sweating and found that atropine was capable of reducing the weight loss of infants due to insensible perspiration 22%, as measured with a Sauter balance. Methylatropine is believed to have equivalent effect.

Effect on the Body Temperature. Even small doses of atropine frequently produce fever without other evidence of toxicity. White[100] (1929) studied this phenomenon and stated that at least 10% of the infants would develop fever of 38° or more when given therapeutic doses of atropine. There has been much argument as to whether or not this is due to the depression of sweating, to increased restlessness or to a direct stimulation of a heat-regulating center in the central nervous system. It has been adequately demonstrated that the hypothalamus is intimately concerned with the mechanism of heat production and heat loss, and that the control can be influenced by drugs (Masserman,[69] 1937, studying antipyretic drugs). Anterior hypothalamus lesions result in hyperthermia.

Use of methylatropine clinically in hypertrophic pyloric stenosis has produced similar results. This probably should not be considered a toxic reaction and is not considered a contraindication for either drug.

ACTION OF THE ATROPINE GROUP ON THE SYMPATHETIC SYSTEM. Bussell[11a] (1940) reviews the rôle of atropine in relation to adrenalin and the sympathetic system. He cites the work of Meyer[69a] (1906) who discovered that strips of ox carotid in Ringer's solution show no spontaneous rhythm, but that the addition of 6 μg. of adrenalin would cause large contractions which were in turn decreased by half if 1.5 mg. of atropine were added along with the adrenalin. Bussell was able to demonstrate that this antagonism held in perfusion of the dog's leg, the rabbit's ear, the nictitating membrane of the cat's eye, and the blood pressure of spinal cats. Atropine also caused dilatation of the spleen, of the small intestine and a fall in blood pressure in cats anesthetized with ether: all sympathetic medi-

ated actions. Hyoscine showed no similar antagonism to adrenalin. The work of Bergengren[4] (1925) indicates that methylatropine also possesses the ability to antagonize the effect of adrenalin on blood-vessels. Atropine has been shown to cause a partial paralysis of the cervical sympathetics in the rabbit's ear perfusion experiment and of the cervical and thoracic sympathetic in studies on cats. There is no similar antagonism between atropine and posterior pituitary extract. These actions of the atropine series on the sympathetic system are not too surprising, in view of its close chemical relationship to cocaine which is a sympathetic blocking agent.

Miscellaneous Effects of the Atropine Group. These drugs are reported to have some local analgesic effects similar to those of cocaine. Edsall and Means[24] (1914) report that the basal metabolic rate and the respiratory quotient are not affected by these drugs. The figures of Nyman on oxygen consumption incidental to his studies on minute volume would tend to bear out this contention. Atropine as such does not affect the blood sugar level. The consensus seems to be that these drugs do not affect milk secretion but that a portion of the drug is excreted *via* the mammary gland in animals and women. It has been reported by Magaard[65] (1882) that lacrimation is decreased. Leukocytosis has been reported in children after atropine. The *voluntary* muscles of mammals are not affected in concentrations that have been tested to date.

Bodily Mechanisms for Handling Atropine in Various Species. Atropine is absorbed rapidly from the gastro-intestinal tract and also disappears from the peripheral blood within a short while. Approximately one-third of the ingested dose may be recovered in the urine 14 hours afterwards. In man the balance is probably destroyed by hydrolysis to tropine and tropic acid in the liver. Traces are detectable in other secretions including milk and sweat. These drugs readily cross the placental barrier.

Some animals possess enzymes capable of hydrolyzing atropine in the serum, brain, kidney and possibly other tissues in addition to the liver. The rabbit, an animal which is able to eat belladonna leaves without evidence of toxicity and is extremely resistant to atropine poisoning, has a high concentration of atropine esterase in its serum. Comparative studies of various tissues by Bernheim and Bernheim[5] (1938) demonstrated atropine esterase activity in the rabbit liver and serum; guinea pig liver also hydrolyzed atropine and homatropine rapidly. The livers of the cat, rat and dog were able to hydrolyze these drugs slowly. There was no such activity in the kidney, brain or serum of the cat, rat, dog or guinea pig. Human serum was also inactive. This enzyme is not identical with choline esterase or the esterase of simple acids such as acetate. Ethylmandalate is hydrolyzed by tissues which do not hydrolyze homatropine. In all tissues tested homatropine was destroyed more rapidly than atropine. (This problem is discussed in detail by Bernheim,[6] 1942.)

The studies of Pilcher[77] (1934) on atropine tolerance in infants and the studies of Schlossmann[80] (1937) on the relationship between age and the action of atropine do not establish this effect as directly attributable to differences in the esterases. Schlossmann comments upon the fact that infants generally show fewer central nervous system symptoms, more marked narcotic effect and unexplainable variations in resistance to these drugs. There seems to be no clinical basis for such differences. Lindberg[60] (1925) describes his experiences with the action of atropine in 100 infants. One infant was given a dose of 6 mg. without harm at the age of 8 months. There were strong vasomotor reactions in healthy infants and the production of fever even in small

doses. Definite tranquilization is produced by atropine in nervous infants.

Experimentally, rabbits of different ages were injected with atropine sulfate intraperitoneally. The LD50 for rabbits under 1 month of age was 0.225 gm. per kg. body weight; that for rabbits over 1 month of age was 0.35 gm. per kg. Rabbits of different ages showed different effects from poisoning. Thus, in the 1st week of life their response to toxic doses was central nervous system paralysis. From the 2nd to 4th week in life, clonic and tonic convulsions were the prominent symptoms with poisoning. Thus, in the first week of life their response to toxic doses was central nervous system paralysis. From the second to fourth week in life, clonic and tonic convulsions were the prominent symptoms with poisoning. Older rabbits showed an increase in heart rate and respiratory rate followed by respiratory paralysis.

THE TOXICITY OF METHYLATROPINE. As might be expected, the toxic reactions to methylatropine nitrate (eumydrin, metropine) are similar in type to those of atropine from which it is derived. Reports both as to its clinical and its experimental toxicity vary. For example, it has been quoted and requoted in the literature that eumydrin is one-fiftieth as toxic as atropine (see Sollman, *Manual of Pharmacology*, 1942), apparently on the basis of Brown and Frazer's work,[10] in 1869, which was on methylatropine iodide (not nitrate) and on unpublished experiments done by Paul Drucker of Rigshospital, Copenhagen, Denmark, in 1927. Usener[92] (1926) quotes Dresser without reference as follows: Lethal dose of atropine 0.1 mg. per kg. in adults; 0.01 mg. per kg. for children; in animals (species not specified) eumydrin was lethal in doses of 0.1 gm. per kg. Kostin (also cited without reference) says that 0.15 gm. per kg. of eumydrin produced death without convulsion (differing from atropine) by paralysis (similar to curare) in rabbits.

Experiments by Graham and Lazarus[38] (1940) indicate, on the basis of intraperitoneal injections of white mice, that eumydrin is 3 times as toxic as atropine. The LD50 of methylatropine nitrate is 0.11 gm. per kg. body weight, while the LD50 of atropine is 0.32 gm. per kg. body weight. They quote Willberg's figure[102] (1914) for atropine in the mouse, LD50 of 0.3 to 0.4 gm. per kg. body weight, as tending to confirm their result. Fromherz[32] (1937) gives the following results for atropine: lethal doses of atropine in gm. per kg. body weight for the mouse, subcutaneously 0.55 gm., intravenously 0.07 gm., orally 0.4 gm.; for the frog, subcutaneously 1.25 gm.; for the rabbit, subcutaneously 0.5 gm., intravenously 0.05; for the cat, subcutaneously 0.13 gm. He compares and comments on the variability of toxicity in various species, particularly the resistance of rodents to atropine. In man, atropine kills by its effect on the central nervous system and is 100 to 500 times as toxic as it is in rodents.

Dyson[23] (1928) argues that the effectiveness of eumydrin is due to the fact that as a dimethyl quarternary ammonium salt, it diverts the action from the rest of the nervous system to the peripheral nerve endings. This may partly explain the divergent opinion of the toxicity of this drug. Studies of Ing, Dawes and Wajada[50] (1945) on the relative potency per molecule in mydriatics showed eumydrin to be 2.22 times as active as atropine in producing mydriasis in the mouse after intraperitoneal injection, but only $\frac{1}{2}$ to 1 time as effective when applied locally to the cat's eye. This difference in mydriatic activity is of the same magnitude as the difference in toxicity of the 2 drugs for the mouse as described by Graham and Lazarus[38] (1940). Death from eumydrin in the Graham experiments was by general paresis and respiratory failure following an initial increase in respiratory rate. No convulsions were observed.

Ing *et al.*[59] argue that the effect of these alkaloids depends on the method of administration. They drew the conclusion that the quarternary methyl salts of the alkaloids (eumydrin) are more active than corresponding tertiary bases (atropine), but that onium salts penetrate all membranes more slowly than tertiary bases, which exist not only as salt cations but also as free base. The effect of age, the rôle of neutralizing agents in the serum, and optical isomerism (eumydrin is racemic) have not been specifically studied as they have been for atropine (Pilcher,[77] 1934; Schlossman,[80] 1937; Cushny,[18] 1920; and others).

Clinically, methylatropine nitrate has been given to infants with hypertrophic pyloric stenosis over periods of weeks with only occasional tonic reactions. Lindberg's[60] (1925) studies on the elimination of atropine probably should hold fairly well for eumydrin. Most of the drug is eliminated in the urine, and the state of hydration, or rather dehydration, was a factor in practically all toxic reactions reported. 'One death with hyperpyrexia was reported by Friedlander[31] (1934) and was attributed to idiosyncrasy. Monrad[71] (1928), using a relatively large dose of 3.5 mg. per day, had 9 toxic reactions in 64 cases, consisting of nystagmus, dilated pupils, fever, flushing, restlessness, and convulsions.' Wallgren[95] (1940) described a flushing of the face as a reliable sign of early toxicity when using alcohol eumydrin applied to the tongue. Mackay[64] (1941) felt that abdominal distention was an important toxic sign and should be watched for. Fifteen of her 40 cases showed mild reactions with doses under 3 mg. per day. Reactions consisted of slight fever, rapid pulse, increased respiration rate, dryness of the mouth, bright red flush and dilated pupils. Vertue[94] (1939) reported no toxic symptoms in 21 cases, using doses of 1.25 mg. per day for 11 weeks. One of Dobbs'[21] (1939) 20 cases showed flushing when getting 4 mg. per day. Braithwaite[8] (1938) and Findlay[30] (1938) discuss 27 cases without evidence of toxicity in dosages of about 3.0 mg. per day. Svensgaard[85] (1935) reports only a few transient reactions as evidenced by clear flaming red skin and a fever of 38° while treating 61 cases with an average dose of 1.75 mg. per day of eumydrin. All reactions, except the death reported by Friedländer[31] (1934), were transient and cleared up when drug was omitted for a short time.

Relation of Dosage to Activity. What then are effective doses of atropine? Henderson[45] (1923) gives some pertinent information on this point. He studied the sensitivity of various nerve endings to atropine and found that the endings (response) of the bulbosacral autonomic outflow are rendered ineffective in the following order: cardiac vagus, chorda secretor, chorda vasodilators, intestinal vagus and bladder. They are depressed in the following order in a 10 kg. dog: (1) 0.1 mg. depressed the chorda secretor from 15 to 10 drops of saliva. (2) 0.2 mg. rendered the cardiac vagus less effective and 0.5 mg. eliminated vagus activity with a consequent increase in heart rate. (3) 0.1 to 0.2 mg. caused a distinct decrease in the tone of the pylorus and small intestine. (4) 0.2 mg. caused a decrease in bladder tone while 0.3 eliminated bladder tone due to vagus. (5) 0.5 mg. dilated the pupil but it was still reactive to light and convergence; maximum dilatation was secured with 0.6 mg. (6) 0.7 mg. caused a slight decrease in blood flow from the salivary gland on chorda stimulation as compared with before atropine. (7) With 30 mg., vagus stimulation produced little if any increase in the rhythmic movements of the small intestine while peristalsis was slight or absent. (8) Doses of 100 mg. caused no decrease in the isotonic or isometric contractions of the bladder on sacral stimulation.

Doses of atropine in a 65 kg. man pro-

duced the following effects: 0.2 mg. caused a decrease in nasal secretion; 0.3 mg. caused dryness of the mouth and relieved griping from constipation; the pupil was not affected. A dose of 2 mg. caused an increase in heart rate to 150 and dilatation of the pupil. Methylatropine is described as having equivalent peripheral effects or possibly stronger curare-like activity than atropine. Yet as with any drug, the effective dose is the dose necessary to produce the desired therapeutic effect. These drugs can only depress or increase the normal activities of various susceptible cells. The clinical impression of a great variability in the response that various individuals show to these drugs is amply confirmed in the wide variability found in the laboratory by the pharmacologists.

Summary. 1. The chemical structure, pharmacologic actions, toxicity and therapeutic uses of atropine, methyl atropine nitrate and related compounds are discussed.

2. The most marked difference between the 2 first-named compounds is in their effects on the central nervous system. Atropine produces marked psychic side effects even in the usual therapeutic range of dosages, whereas methyl atropine does not. Death from atropine is preceded by convulsions. Death from methyl atropine is due to paralysis and failure of respiration without convulsions.

3. The peripheral actions of the 2 compounds are apparently similar, but the responses vary with the route of administration. Methylatropine has the weaker action in equivalent doses when applied *topically* as in ophthalmic use. Apparently, *oral* potency is approximately equal, although some investigators state that atropine is 3 or 4 times stronger. *Parenterally* methylatropine is probably somewhat stronger than atropine.

4. The essential action of both compounds is antivagal and they apparently prevent effector cells from reacting to acetylcholine.

5. The compounds are discussed in some detail in relation to each system of the body as follows: eye, respiratory tract, heart, salivary gland, esophagus, stomach, pylorus, small intestine, colon, smooth muscle, biliary duct tract, gall bladder, uterus, urinary bladder, sweat glands, and sympathetic nervous system.

6. The animal body probably detoxifies these compounds by hydrolysis to tropine and tropic acid. The existence of atropine esterases has been confirmed in many animal tissues. This enzyme may be unable to act on methylatropine.

7. It has long been repeated that methylatropine is only one-fiftieth as toxic as atropine. By intraperitoneal injection recent experimental evidence shows it to be 3 times as toxic for white mice as is atropine.

8. The principal clinical use to date of methylatropine has been in hypertrophic pyloric stenosis and in infantile pyloric spasms.

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BOOK REVIEWS AND NOTICES

HISTOPATHOLOGIC TECHNIC. By R. D. LILLIE, M.D., Medical Director, U. S. Public Health Service. Pp. 300. Phila.: Blakiston, 1948. Price, $4.75.

THIS book should prove of value to all who are concerned with the preparation of tissue sections. The author admittedly does not attempt to present an encyclopedia of tissue technique, but rather is concerned with what in his opinion are the best methods for each particular purpose. The book is conveniently arranged so that stains for the various histologic structures are grouped together. In addition to the standard procedures given in most other books on the subject, there are excellent chapters on the staining of enzymes and pigments. These and many other special techniques, which are frequently neglected in laboratory manuals, are readily available in this book.

A. R.

SEXUAL BEHAVIOR IN THE HUMAN MALE. By ALFRED C. KINSEY, Prof. of Zoology, WARDELL B. POMEROY, Research Assoc., and CLYDE E. MARTIN, Research Assoc., Indiana Univ. Pp. 804. Phila.: W. B. Saunders, 1948. Price, $6.50.

THIS book is one of the most important recent contributions to the study and understanding of the fundamental biologic problem of sex in man. Sexual histories of 5300 males have been thoroughly analyzed, and are objectively and statistically evaluated. This male group represents a very adequate cross-section of our male population, as it includes large numbers from nearly all educational and social levels. The text includes brief reviews of results of other workers in this field, a study of the statistical methods used, and techniques involved in obtaining an adequate sexual history. Variations in sexual activity with social and educational classes, age and religious background are correlated and explained. Practically all possible sources of sexual outlet are considered with respect to incidence in various groups, techniques and social implications.

The problem of male sexual behavior has been soundly and keenly attacked from all angles; the results obtained—unbiased and objective statistics—are very instructive and occasionally revelational.

I. Z.

THE PATHOLOGY OF TRAUMATIC INJURY. A GENERAL REVIEW. By JAMES V. WILSON, M.D., M.R.C.P. (Lond.), Pathologist to Harrogate and District General and Royal Bath Hosps., D.A.D.P., Malta Command, 1940–1943. Pp. 192; 61 ills. Balt. and London: Williams & Wilkins, 1946. Price, $6.00.

THIS small volume is based on the author's experience with the Royal Army Medical Corps in the Second World War. Intended as a general review, it aims to "mention progress and at the same time outline difficulties and problems awaiting urgent solution, to deal with essentials rather than give a mass of irrelevant facts."

Though written abroad, under difficulties, and drawing its material chiefly from the traumata incident to modern war, the book for the most part achieves its goals. Best covered are the effects of blast, the rediscovered "crush syndrome" and the increasingly recognized embolism of fat. There is a chapter on burns, but effects of war gases and atomic weapons are not considered. A modest number of references up to 1946 serve as a basis for modern ideas.

The predominantly surgical viewpoint, with many pages of advice on diagnosis and treatment, is distracting in a book with this title. The surgeon's inferences on therapy may be quite different from those of the pathologist, who is not responsible for the care of injured patients. It is hoped this point will be given consideration in a later edition.

C. B.

NUCLEIC ACID. SYMPOSIA OF THE SOCIETY FOR EXPERIMENTAL BIOLOGY. No. I. Edited by J. F. DANIELLI and R. BROWN. Pp. 290; 18 ills. Cambridge, at the University Press: New York: Macmillan, 1947. Price, $8.50.

THIS book is a collection of papers read at a symposium in July, 1946. The various experimental approaches to the study of nucleic acids are considered and evaluated by such eminent workers as Danielli, Caspersson, Brachet, Darlington, Gulland, and many others. The biochemical structure and methods of synthesis of nucleis acids and their purine derivatives are thoroughly reviewed. The various histochemical techniques used for the identification of nucleic acids are described and criticized in detail. Among other studies of nucleoproteins are included histochemical observations on tumor tissue, the relationship of nucleoprotein metabolism to nerve function, *x*-ray studies and effects, and the function of nucleoprotein in the cell nucleus. Most of the papers treat comprehensively one or several aspects of nucleoprotein study and the results achieved. Knowledge of nucleoproteins is thus effectively concentrated in this excellent text. I. Z.

DISEASE OF THE NOSE, THROAT AND EAR. By WILLIAM LINCOLN BALLENGER, M.D., F.A.C.S., Late Prof., Univ. of Illinois School of Medicine, and HOWARD CHARLES BALLENGER, B.S., M.D., F.A.C.S., Assoc. Prof. of Otolaryngology, Northwestern Univ. School of Medicine. Assisted by JOHN JACOB BALLENGER, B.S., M.D. 9th ed. Pp. 993; 597 ills.; 16 plates. Phila.: Lee & Febiger, 1947. Price, $12.50.

The latest revision of this standard work brings the text and illustrations up to date. New chapters dealing with headache, neuralgias of the face and head, and rhinoplastic surgery have been added. Authoritative revisions of the section on bronchoscopy, laryngoscopy, esophagoscopy and gastroscopy have been carried out by Gabriel Tucker and C. L. Jackson, while those on the physiology and functional tests of the labyrinth and inflammatory diseases of the labyrinth were contributed by Alfred Lewy.

Embracing the entire field of otolaryngology, the book employs the orthodox method of approaching each disease by first presenting the fundamental anatomical, physiological and pathological considerations, and then discussing etiology, symptoms, diagnosis and treatment. The therapeutic suggestions are those which meet the acid tests of time and practice. The authors have appreciated the instruction value of illustrating the instrumentarium for various surgical procedures.

Controversial subjects have been avoided. A frank discussion of a few of these, such as the present role of vitamin therapy in otolaryngology, might have been profitably introduced as a guide to students and general practitioners. Most of the bibliographical references are arranged at the foot of the page so as to be readily accessible. The value of a reference volume of this scope and magnitude, however, would be enhanced by a more complete bibliography.

This work, both comprehensive and condensed, continues to be a leader in its field and invaluable to the medical student, the general practitioner and the otolargyngologist. H. S.

UNIPOLAR LEAD ELECTROCARDIOGRAPHY. By EMANUEL GOLDBERGER, B.S., M.D., Clinical Lecturer in Medicine, Columbia University, Faculty of Medicine. Pp. 182; 88 ills. Phila.: Lea & Febiger, 1947. Price, $4.00.

This monograph is the result of several years' study to determine the practicability of routine clinical use of unipolar leads and the correlation of unipolar extremity leads, precordial leads and the standard leads. The author is well qualified by training and experience to present this subject. The approach to the interpretation of the electrocardiogram is made in terms of basic unipolar lead patterns, rather than standard limb lead patterns, as is the rule in most texts.

The theoretical considerations of the clinical use of unipolar leads are based largely on the Einthoven triangle theory. The author advocates the routine use of 6 unipolar precordial leads, 3 unipolar extremity leads (or "augmented" unipolar extremity leads), and the usual limb leads. Special emphasis is placed on the variations in the unipolar extremity leads and standard limb leads which occur with changes in position of the heart. The author believes that the use of unipolar leads is of particular advantage in the following conditions: The interpretation of axis deviation; the diagnosis of Q wave abnormalities, especially Q3 waves; the differentiation of pulmonary embolism and posterior infarction; the diagnosis of small or multiple infarcts; the differentiation of bundle branch block; the diagnosis of right ventricle hypertrophy, and the interpretation of abnormalities of the RS-T segments and T waves.

Evidence is offered in support of the au-

thor's theses, and the illustrations are adequate and clear. (Incidentally, an error was noted in the correlation of the text with Figure 41B.) The theoretical aspects of the subject are presented in a manner that can be grasped by the average physician. Some of the interpretations of patterns obtained from ventricular hypertrophy with variations in the position (and rotation) of the heart may be questioned by other authorities. The last word has not been spoken on many phases of the subject, and one must agree with the author when he says "the significance of many electrocardiographic changes is still obscure, even in the unipolar leads." The monograph is a distinct contribution to the field of electrocardiography and should be read with interest and profit by all cardiologists and internists. J. V.

OFFICE TREATMENT OF THE EYE. By ELIAS SELINGER, M.D., Attending Ophthalmologist, Mount Sinai, Cook County and Michael Reese Hosps. Pp. 542; 67 ills. Chicago: Year Book Publishers, 1947. Price, $7.75.

This book is a very practical handbook of treatment intended for practicing opthalmologists. There are excellent sections on the general aspects of chemotherapy, errors of refraction and the usual diseases of the eye and its adnexa. It is pleasing to see that where modern treatment has effectively replaced older procedures, this book recognizes the fact by omitting them. The illustrations are excellent. F. A.

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400 YEARS OF A DOCTOR'S LIFE. Collected and Arranged by GEORGE ROSEN, M.D., and BEATE CASPARI-ROSEN, M.D. Pp. 429. New York: Henry Schuman, 1947. Price, $5.00.

The authors have achieved an interesting experiment in biographical montage. For this anthology of the arc and the stations of the physician's existence, they drew on "more than eighty personalities (four of them women) who have made significant contributions to the literature of medical autobiography."

The organization of the text is unique. Under a dozen such headings as Early Years; Scientist; Scholar; Teacher; The Doctor as Patient; The Doctor Goes to War; Writing and Politics, the authors collate from several different sources what each of the personalities had written on the particular score. This arrangement affords us the memories of their early years, as reported by such divergent authors as Daniel Drake, Havelock Ellis and Santiago Ramon y Cajal, and the reflections on life and death of the 21 year old Virchow, the despairing Horace Wells, the life-wise Walter Cannon, and the stoical Hans Zinsser.

The effect of such collations is at times puzzling and provocative, but always interesting. The one conviction that emerges is that there is no medical prototype, no "homo medicus." Physicians differ in origins, in constitutional and personality types, in experiences, and in all else, as widely and as ardically as do all men in all things. Save perhaps for their devotion to their profession, there is no distinguishing common denominator among doctors. The sum of the personal experiences of the "eighty personalities" abstracted in the Rosen anthology does not, to this reviewer's mind, yield a "composite autobiography of The Doctor".

But the Rosen anthology yields more real and more valuable assets. It provides good reading, both in the biographical notes which precede each citation, and in the citations selected. It provides good sampling, to inform and to sharpen one's interest in distinguished men and women who have served in the most humane and compassionate of professions. It is a fine volume for random reading, and a fine guide to the best in medical autobiography. I. G.

BLOOD DERIVATIVES AND SUBSTITUTES. By CHARLES STANLEY WHITE, M.D., Sc.D., Former Prof. of Surgery, George Washington Univ. School of Medicine, and JACOB JOSEPH WEINSTEIN, B.S., M.D., Assoc. in Surgery. Pp. 484; 190 ills. Balt.: Williams & Wilkins, 1947. Price, $7.50.

THE title of this book of 15 chapters amply summarizes the scope of the contents. Following an introductory historical chapter, some aspects of the chemistry and physiology of plasma are presented, with much of the space given to commonly employed laboratory methods. Chapters dealing with preparation, storage and administration of plasma, plasma fractions and by-products, whole blood and substitutes, are complete in details and generously illustrated. The last third of the book deals with the etiology and treatment of shock, and the results of plasma therapy. Throughout the book points of clinical interest are usually illustrated with case reports.

Most of the facts required for operating a

blood and plasma bank and the principles necessary for their intelligent use are to be found. One questions, however, the need for including laboratory procedures which may be found in any book of laboratory technique. A serious deficiency is the absence of an adequate treatment of blood groups and the Rh factor. Under the heading of "Infectious Jaundice" reports on the apparent transmission of jaundice by blood or plasma are cited (p. 463), but the importance of homologous serum jaundice has apparently escaped the authors since they do not discuss the desirability of using unpooled plasma. R. N.

INFANT NUTRITION. By P.C. JEANS, A.B., M.D., Prof. of Pediatrics, College of Medicine, State Univ. of Iowa, and WILLIAMS MCKIM, MARRIOTT, B.S., M.D., Late Prof. of Pediatrics, Washington Univ. School of Medicine. 4th ed. Pp. 495; 36 ills. St. Louis: C. M. Mosby, 1947. Price, $6.50.

THIS text contains in very readable form a good combination of fundamental nutritional information together with many practical points for the application of such information. The facts have been brought up to date and are presented in a thoroughly sound and effective way. K. E.

INTERNAL MEDICINE IN GENERAL PRACTICE. By ROBERT PRATT MCCOMBS, B.S., M.D., F.A.C.P., Asst. Prof. of Medicine, Tufts College Medical School, etc. 2nd ed. Pp. 741; 122 ills. Phila., and London: W. B. Saunders, 1947. Price, $8.00.

THE author has attempted to present a short, practical book on internal medicine for the general practitioner, stressing particularly methods of diagnosis. This edition contains much useful information in the form of tables of differential diagnosis and indications for and techniques of important diagnostic aids. It is up to date and embodies a thorough and thoughtful approach to the problems of internal medicine. Many practical hints are scattered throughout. The arrangement of the material is good and the amount of space allotted to the various diseases corresponds more nearly to their incidence in practice than in the usual text book of medicine.

Turning to shortcomings, in a few instances, such as the use of digitalis in congestive heart failure, the methods of administration of a drug are not specific enough. In some diseases, such as ulcerative colitis and glomerulonephritis, not enough information as to the clinical course is given to permit intelligent management.

In general, however, this revised book should prove a useful addition to the library of the practitioner of general medicine for whom it was intended. It also has much to recommend it as a book for the fourth year medical student or intern. A. R.

HANDBOOK OF PSYCHIATRY. By WILLIAM OVERHOLSER, M.D., Prof. of Psychiatry, George Washington Univ., and WINIFRED V. RICHMOND, M.D., Late Chief, Department of Psychology, St. Elizabeth's Hosp. Pp. 252. Phila.: J. B. Lippincott, 1947. Price, $4.00.

PSYCHOLOGY and psychiatry are interwoven in this book in a refreshing and sincere manner. The authors have put the various types of human behavior and motive into terms comprehensible by the average college student, meanwhile accomplishing a hitherto unattempted correlation of the several "schools of thought" of psychiatry with psychology, general medicine, and allied sciences.

The chief source of clinical material was St. Elizabeth's Hospital, which has actively treated patients from all of the United States and its possessions. This wealth of clinical material comes into the text in the form of abstracts of case histories, skillfully prepared and selected.

While the book as a whole is outstanding, the chapters dealing with the modern mental hospital, mental aberrations of war, forensic psychiatry, child psychiatry and "psychiatry and the layman" are especially recommended. The fourth chapter, dealing with mental deficiency, despite confusing use of terminology, is unparalleled in similar texts.

The authors acknowledge that crusading laymen, following the example of Dorothea Lynch Dix, to whom this book is dedicated, were largely responsible for and should be credited with much of the early advances in the field of psychiatry. They hope that "in the campaign for the development of better human beings, for raising the level of our human resources" psychiatrists may depend upon the backing of a body of informed public opinion, without which psychiatrists would be "as helpless as a general without an army."

There is an excellent bibliography.

E. B.

NEW BOOKS

The Sulfonamides and Allied Compounds. By ELMORE H. NORTHEY, PH.D., American Chemical Monograph Series. Pp. 660. New York: Reinhold, 1948. Price, $12.50.

Occupational Medicine and Industrial Hygiene. By RUTHERFORD T. JOHNSTONE, M.D., Consultant in Industrial Health, Lecturer, Univ. of California. Pp. 604; 117 ills., 7 in color. St. Louis: C. V. Mosby, 1948. Price, $10.00.

Tuberculosis. By FRANCIS MARION POTTENGER, M.D., F.A.C.P., Emeritus Prof. of Medicine, Univ. of Southern California. Pp. 598; 105 ills. St. Louis: C. V. Mosby, 1948. Price, $12.00.

Brief Psychotherapy. By BERTRAND S. FROHMAN, M.D., with the collaboration of EVELYN P. FROHMAN. Foreword by WALTER C. ALVAREZ, M.D. Pp. 265. Phila.: Lea & Febiger, 1948. Price, $4.00.

Proteins and Amino Acids in Nutrition. Edited by MELVILLE SAHYUN, M.A., PH.D. Pp. 566. New York: Reinhold, 1948. Price, $7.50.

The Rh Factor in the Clinic and the Laboratory. Edited by JOSEPH M. HILL, M.D. and WILLIAM DAMESHEK, M.D., Special, Issue No. 2 of Blood, *The Journal of Hematology.* Pp. 192. New York: Grune & Stratton, 1948. Price, $4.25.

Deep Analysis. By CHARLES BERG, M.D. (LOND.), D.P.M., Fellow of the British Psychological Association. Pp. 254. New York: W. W. Norton, 1947. Price, $3.50.

Atlas of Bacteriology. By R. CRANSTON LOW, M.D., F.R.C.P.E., F.R.S.E. (EDIN.), and T. C. DODDS, F. I. M. L. T. 168 ills., 167 in color. Balt.: Williams & Wilkins, 1947. Price, $8.50.

Cardiopatias Congenitas de la Infancia. By DR. AGUSTIN CASTELLANOS Y GONZALEZ. Pp. 406; 20 ills. Havana, Cuba: M. V. Fresneda, 1948. Price, $9.00.

Diabetes Mellitus in General Practice. By ARTHUR R. COLWELL, M.D., Assoc. Prof. of Medicine, Northwestern Univ. Pp. 350. Chicago: The Year Book Publishers, 1947. Price, $5.25.

Diseases of the Joints and Rheumatism. By KENNETH STONE, D.M. (OXON.), M.R.C.P Pp. 362; 58 ills. New York: Grune & Stratton, 1947. Price, $6.50.

Experimental Air-borne Infection. By THEODOR ROSEBURY, with the co-authorship of the Staff of the Laboratories of Camp Detrick, Md. Pp. 222; 40 figs. Balt.: Williams & Wilkins, 1947. Price, $4.00.

Applied Medical Bacteriology. By MAX S. MARSHALL, PH.D., with the collaboration of JANET B. GUNNISON, M.A., ALFRED S. LAZARUS, PH.D., ELIZABETH L. MORRISON, M.A., and MARIAN C. SHEVKY, A.B., Medical Center of the Univ. of California. Pp. 340; 8 ills. Phila.: Lea & Febiger, 1947. Price, $4.50.

The Pathology of Nutritional Disease. By RICHARD H. FOLLIS, JR., M.D., Assoc. Prof. of Pathology, Duke Univ. School of Medicine. Pp. 291; 71 ills. Springfield, Ill.: Charles C Thomas, 1947. Price, $6.75.

Advances in Military Medicine. Science in World War II. Edited by E. C. ANDRUS, D. W. BRONK, G. A. CARDEN, JR., C. S. KEEFER, J. S. LOCKWOOD, J. T. WEARN, W. C. WINTERNITZ, and TUCKERMAN DAY. Foreword by ALFRED N. RICHARDS. In 2 vols. Pp. 900; 94 figs. Boston: Little, Brown, 1948. Price, $12.50.

The Biology of Melanomas. Edited by ROY WALDO MINER, MYRON GORDON, and LOTHAR SALIN. Vol. IV. Pp. 478; 107 plates. New York: The New York Academy of Sciences, 1948. Price, $5.00. To members, $4.00.

Blue Cross and Medical Service Plans. By LOUIS S. REED. Pp. 323. Washington, D. C.: U. S. Public Health Service, 1947.

Telepathy and Medical Psychology. By JAN EHRENWALD, M.D. Pp. 212. New York: W. W. Norton, 1948. Price, $3.00.

Case Histories in Clinical and Abnormal Psychology. Edited by ARTHUR BURTON, Assoc. Prof. of Psychology, Willamette Univ., and ROBERT E. HARRIS, Assoc. Prof. of Medical Psychology, Univ. of California. Pp. 680. New York: Harper & Brothers, 1948. Price, $4.00.

NEW EDITIONS

Disability Evaluation. By EARL D. MCBRIDE, M.D., F.A.C.P., Diplomate, American Board of Orthopedic Surgery. 4th ed. Pp. 667; 400 ills. Phila.: J. B. Lippincott, 1948. Price, $12.00.

The Treatment of Rheumatism in General Practice. By W. S. C. COPEMAN, O.B.E., M.A., M.D. (CANTAB.), F.R.C.P.–LOND.). 4th ed. Pp. 258. Balt.: Williams & Wilkins, 1946. Price, $4.00.

Communicable Disease Control. By GAYLORD W. ANDERSON, M.D., DR.P.H., Mayo Prof. and Director, School of Public Health, Univ. of Minnesota, and MARGARET G. ARNSTEIN, R.N., M.A., M.P.H. 2nd ed. Pp. 450. New York: The Macmillan Company, 1948. Price, $5.00.

Endotracheal Anæsthesia. By NOEL A. GILLESPIE, M.A. (OXON.), M.D. (WIS.), Assoc. Prof. of Anæsthesia, Univ. of Wisconsin. 2nd ed. Pp. 237; 56 figs; 1 color plate. Madison: University of Wisconsin Press, 1948. Price, $4.00.

Treatment by Diet. By CLIFFORD J. BARBORKA, M.D., D.Sc., F.A.C.P., Asst. Prof. of Medicine, Northwestern Univ. Medical School. 5th ed. Pp. 784; 14 ills., 13 in color. Phila.: J. B. Lippincott, 1948. Price, $10.00.

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